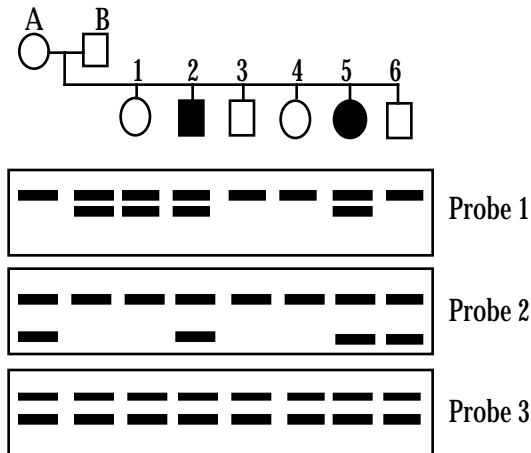


1. Couple A and B have 2 children with Tay-Sachs disease and 4 children who are unaffected by the disease. RFLP analysis was performed on the family using 3 different probes made from sequences known to be closely linked to the gene responsible for Tay-Sachs disease.



a. (2 pts) Which probe(s) can be used to deduce genotypes in this family? **1 and 2**

b. (4 pts) Which of the unaffected children have inherited a disease allele, and from which parent did they inherit it?

Child 1 - inherited from father (B)
Child 6 - inherited from mother (A)

c. (4 pts) If child 6 grows up and marries a woman whose sister had Tay-Sachs disease, what is the probability that they will have an affected child?

Child 6 is heterozygous, so probability of an affected child will be probability of his wife being heterozygous (2/3) x probability of homozygous recessive offspring (1/4) = 1/6

2. (4 pts) In a Neurospora linear ascus, which of the following spore ratios would indicate that DNA mismatch repair has NOT occurred. (Please indicate all correct answers)

- A. 2:6, 6:2, 3:5, 5:3, and 4:4
- B. 2:6, 6:2, 3:5, and 5:3
- C. 2:6, 6:2, and 4:4
- D. 2:6 and 6:2
- E. 3:5 and 5:3**
- F. None of the above

3. (4 pts) SINE elements in humans and Ds elements in corn have in common which of these features (Please indicate all correct answers):

- A. Both move replicatively.
- B. Both move non-replicatively.
- C. Both require the presence of another element in the genome to move.**
- D. Both are similar to retroviruses.
- E. Neither encodes a functional gene product.**
- F. Both can disrupt gene function in the host upon insertion.**

G. None of the above.

4. (15 pts) In the following list of proteins, there are pairs that bind directly to each other to form a complex. List **5 pairs** and for each pair, choose all the numbered statements that are true for that complex (any statement can be used more than once).

- | | | | |
|----------|------------------|---------------|---------------------------------|
| cyclin B | securin | E2F | c-jun |
| c-fos | ubiquitin ligase | Cdk | separase |
| RB | cyclin D | cycloheximide | destruction box binding subunit |

a. Cyclin B		+ Cdk	(1) regulated by phosphorylation of a subunit
Properties (please write numbers only): 2, 3, 6			(2) regulated by ubiquitination of a subunit
b. Cyclin D		+ Cdk	(3) causes phosphorylation of a different protein
Properties: 2, 3, 5			(4) causes ubiquitination of a different protein
c. separase		+ securin	(5) controls entry into S phase
Properties: 2, 8			(6) controls entry into M phase
d. ubiquitin ligase		+ destruction box binding subunit	(7) controls apoptosis
Properties: 1, 4, 8			(8) controls completion of mitosis (anaphase)
e. RB		+ E2F	(9) controls transcription of cell cycle genes
Properties: 1, 5, 9			Also: c-fos + c-jun (~1) 5, 9

5. (4 pts) Colorblindness is an X-linked trait in humans. Two brown-eyed parents with normal color vision have a color blind, blue-eyed child. Assuming that blue/brown eye color is inherited autosomally, which of the following statements is true (Please indicate all correct answers):

- A. The child has to be male.**
- B. The child has to be female.
- C. The sex of the child cannot be deduced from this information.
- D. One parent is homozygous for the brown allele.
- E. Both parents are heterozygous at the eye color locus.**
- F. The father is the source of the colorblindness allele.

G. The mother is the source of the colorblindness allele.

H. None of the above.

6. HeLa cells are cancer cells derived from an epithelial tumor removed from a patient (Henrietta Lacks) in the 1950's. Her tumor cells have continued to proliferate rapidly in tissue culture dishes around the world, long after she died.

a. (2 pts) If you measured telomerase activity in HeLa cells would you expect to find high or low levels, relative to normal epithelia (somatic) cells?

HIGH

b. (4 pts) How does the difference in telomerase levels affect the behavior of cancer cells compared with normal somatic cells?

High telomerase in cancer cells allows for "immortality" = unlimited rounds of replication. Low (absent) telomerase in somatic cells limits the number of rounds of replication, resulting in apoptosis when the telomeres shorten to a critical length.

7. Since HeLa cells grow well in culture dishes, they make a good model system to look for mutations in cell division machinery. Suggest a likely mutation for each of the following mutant phenotypes, Be as specific as possible, indicating both the gene and specific mutation (knock out mutation, specific amino acid change, etc.).

a. (2 pts) Failure to condense chromosomes in prophase. The cells have otherwise normal prophase (mitotic spindle forms, nuclear envelope breakdown, etc.).

mutant gene: HISTONES(H1) CONDENSIN	specific mutation: MISSENSE = PREVENTS PHOSPHORYLATION BY MPF KNOCK OUT OR ANY LOSS OF FUNCTION MUTATION
---	--

b. (2 pts) Failure of the nuclear envelope to breakdown in prophase. The cells have otherwise normal prophase (mitotic spindle, chromosomes condense, etc.).

mutant gene: NUC. LAMIN	specific mutation: MISSENSE = PREVENTS PHOSPHORYLATION BY MPF
-----------------------------------	---

c. (2 pts) Failure to assemble mitotic spindle in prophase. Centrosomes are replicated in S-phase and the cells have otherwise normal prophase (chromosomes condense, nuclear envelope breakdown, etc.).

mutant gene: MAP4	specific mutation: MISSENSE = PREVENTS PHOSPHORYLATION BY MPF
-----------------------------	---

8. (4 pts) Which of the following processes can contribute to the **spontaneous** mutation rate? (Please indicate all correct answers)

A. base intercalation

B. deamination

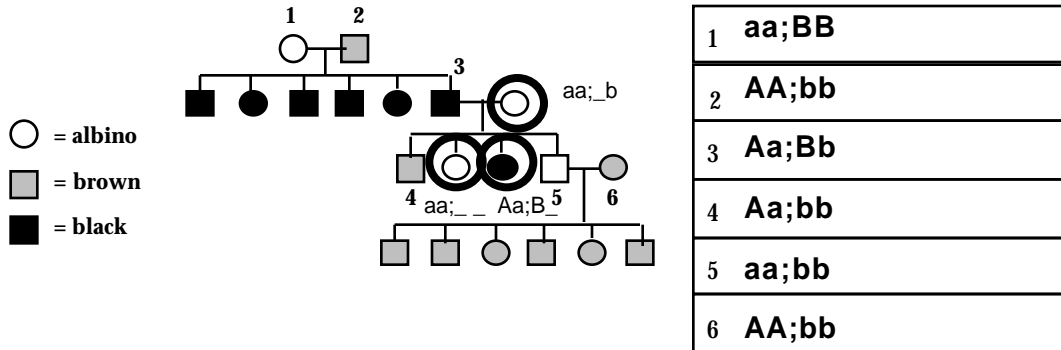
C. tautomerization

D. DNA polymerase errors

E. SOS response

F. None of the above.

9. In the following dog pedigree, use A/a for alleles of the albino gene and B/b for alleles of the melanin color gene to state the genotypes of the indicated individuals (6 pts).



(2 pts) Circle all the dogs in this pedigree for whom the exact genotype cannot be deduced.

10. (4 pts) A colorblind man with Klinefelter's syndrome has parents with normal color vision. Which of the following statements can explain this man's genotype? (Please indicate all correct answers)

- A. The mother's sex chromosomes suffered non-disjunction in the first meiotic division.
- B. The mother's sex chromosomes suffered non-disjunction in the second meiotic division.**
- C. The father's sex chromosomes suffered non-disjunction in the first meiotic division.
- D. The father's sex chromosomes suffered non-disjunction in the second meiotic division.
- E. The mother's sex chromosomes suffered non-disjunction in either the first or the second meiotic division.
- F. The father's sex chromosomes suffered non-disjunction in either the first or the second meiotic division.
- G. None of the above.

11. You have discovered a yellow-flowered mutant in pea plants. When it is crossed to what you believe is a wild-type pea plant, all of the F1 progeny have the wild-type purple flower color. When these F1 plants are self-crossed, you observe 1260 purple-flowered plants and 340 yellow-flowered plants.

a. (6 pts) Do these numbers deviate significantly from the expected monohybrid ratio? Please explain.

Yes, these numbers deviate significantly from a 3:1 monohybrid ratio. This can be calculated using the chi square equation, which sums the square of observed - expected divided by the expected. For 1600 total progeny, we would expect 1200 to show the dominant purple flower phenotype and 400 to show the homozygous recessive yellow flower color.

$$\text{Chi square} = \frac{(1260 - 1200)^2}{1200} + \frac{(340 - 400)^2}{400} = \frac{(60)^2}{1200} + \frac{(60)^2}{400} = \frac{3600}{1200} + \frac{3600}{400} = 3 + 9 = 12$$

There is one degree of freedom, because there are only two phenotypic classes. With one degree of freedom, you would expect to see a deviation of 12 less than 0.5% of the time. Chi square values that indicate a deviation that occurs less than 5% of the time are considered statistically significant.

Therefore these numbers deviate significantly from the expect 3:1 monohybrid ratio. The numbers actually reflect a 13:3 ratio with statistical significance.

b. (4 pts) Which of the following possibilities might explain the observed F2 phenotypic ratio. (Please indicate all correct answers)

- A. The yellow-flowered phenotype is epistatic to the purple-flowered phenotype.
- B. The supposedly wild-type pea plant carried a recessive suppressor mutation.**
- C. The supposedly wild-type pea plant carried a dominant suppressor mutation.

- D. The yellow-flowered mutation is an unstable allele that reverts somatically.
E. The supposedly wild-type pea plant carries a deletion that removes the yellow-flower gene.
F. None of the above.

12. A white-eyed female fruitfly from a pure-breeding white-eyed stock is crossed to a wild-type red-eyed male fly. All of the F1 progeny have wild-type red eyes. When an F1 female fly is mated to a white-eyed male from the pure-breeding stock, the following phenotypic ratios are observed:

300	white-eyed	
300	red-eyed	= 600 parental
200	orange-eyed	
200	brown-eyed	= 400 recombinant between
<u>1000</u>		<i>cn</i> and <i>bw</i> mutations = 40 m.u.

a. (2 pts) Do these data indicate X-linked or **autosomal inheritance**? (Circle one)

b. (2 pts) How many genes are involved in the eye color phenotypes described here? (Circle one)

0 1 2 3 4 5

c. (6 pts) Which of the following statements describe the gene or genes involved (Please indicate all correct answers):

- A. single gene with multiple alleles showing complete dominance.
B. single gene with multiple alleles showing incomplete dominance.
C. single gene with multiple alleles showing co-dominance.
D. multiple genes located on different chromosomes.
E. multiple genes located on same chromosome and more than 50 map units apart.
F. multiple genes located on same chromosome, between 25 and 50 map units apart.
G. multiple genes located on same chromosome and between 10 and 25 map units apart.
H. None of the above.

13. When 2 different wild strains of *Neurospora* are crossed to a standard laboratory strain, 50% of the asci are defective, and contain inviable spores. Propose a cause for the spore inviability in each case:

a. (5 pts) In the cross with strain 1, all of the spores within the defective asci are inviable.

This indicates that the wild strain had a reciprocal translocation. In fungi, all of the products of a single meiosis remain together in an ascus. In the translocation heterozygote meiocytes, half of the meiotic events will have adjacent-1 segregation during the first meiotic division, so that each meiotic product receives one translocated and one normal homolog for the two chromosomes involved in the translocation. All of these products (spores) will have gene imbalance and will be inviable. The other half of the meioses will have alternate segregation during the first meiotic division, so that each meiotic product receives either both translocated chromosomes or both normal chromosomes. All of the ascospores in these asci will be viable.

b. (5 pts) In the cross with strain 2, half of the spores within the defective asci are inviable.

This indicates that the wild strain had an inversion involving at least 50 map units on one chromosome. In the inversion heterozygote meiocyte,

recombination within the inversion loop will result in aneuploid meiotic products. The defective asci are those in which recombination within the loop has occurred (50%). In the recombination event, only two of the nonsister chromatids were involved, while the outer sister chromatids were unaffected. Therefore only half of the ascospores will be inviable within these recombinant asci.

14. In the following open reading frame, single transition mutations will create premature stop codons that would destroy protein function.

5'-ATGTTCTGGCTACTGGGACCGCAAAAACCAGGTC.....

a. (4 pts) Which triplet codon would change to a stop codon upon treatment with EMS?

triplet codon:	amino acid encoded:	stop codon induced:
UGG	Trp	UGA or UAG

b. (4 pts) Which triplet codon would change to a stop codon upon treatment with 5-bromouracil?

triplet codon:	amino acid encoded:	stop codon induced:
CAG	Gln	UAG

c. (4 pts) What part of this sequence might be prone to frameshift mutations and why?

The AAAAA repetitive sequence will be prone to slipped mispairing during DNA replication. If the double helix denatures and the nascent strand reanneals with one A buckled out, the new strand will have an insertion of one nucleotide relative to the template strand. Since this is within a coding region, it would cause a frameshift during translation of the mRNA. Likewise, if the nascent strand reanneals with the template strand buckled out, it will have a deletion of a single nucleotide relative to the normal sequence and this will shift the reading frame in the opposite direction.

15. (4 pts) An unstable allele, a1, of the anthocyanin biosynthetic enzyme gene, A, in corn reverts at high frequency in the presence of the Dt allele. What phenotypic ratios would you expect to see in the kernels resulting from a self-cross of A/a1;Dt/dt plants? (Please indicate all correct answers)

A. 9 purple: 6 dotted: 1 white.

B. 9 purple: 3 pink: 3 dotted: 1 white.

C. 9 purple: 3 dotted: 4 white.

D. 12 purple: 4 white.

E. 15 purple: 1 white. 9 A/__;Dt/__ + 3 A/__;dt/dt 3 a1/a1;Dt/__ 1 a1/a1;dt/dt

F. None of the above. (12 purple:3 dotted:1 white)

16. (4 pts) Marfan syndrome is an autosomal dominant human disorder that affects connective tissue. Marfan patients typically have tall stature, elongated faces, and malformed hearts that are prone to aortic rupture.

All individuals carrying the disease allele show some aspect of these phenotypes, but to varying degrees. Which of the following statements is true about Marfan syndrome? (Please indicate all correct answers)

A. The mutation is pleiotropic in its effects.

B. The syndrome shows full penetrance but variable expressivity.

C. The syndrome shows variable penetrance but full expressivity.

D. The syndrome shows variable penetrance and variable expressivity.

E. The syndrome shows full penetrance and full expressivity.

F. None of the above.

17. Genetically engineered corn uses a strong promoter from a gene found in Cauliflower Mosaic Virus (CaMV promoter) to express large amounts of a bacterial protein that makes the corn resistance to insect pests. Critics of genetically engineered corn contend that recombination between the corn and human DNA might introduce the CaMV promoter into the human genome, potentially causing cancer.

a. (4pts) What sequence elements must be present in the CaMV promoter in order for it to activate high levels of gene expression in humans? Be as specific as possible, identifying specific sequence elements and the proteins that bind to them.

- **TATAA Box = binds TFIID (TBP, basal TF's for RNA Pol II)**

- **Proximal promoter elements = binds constitutive TF's**

(- **Enhancers or response elements that bind regulated TF's present in human cells**)

b. (5 pts) How might the accidental joining of a strong viral promoter with a normal human gene cause cancer?

If the viral promoter is joined close to and upstream (5') of the normal transcription unit (transcription start site) then it may drive high levels of constitutive (unregulated) expression of the normal gene.

If the normal gene is a proto-oncogene then the resulting mutant is a an oncogene. Constitutive expression of the oncogene may activate the cell cycle, inducing the unregulated proliferation of the cell to produce a tumor.

Proto-oncogenes include genes that encode components of the growth factor signal transduction pathways (growth factors, receptors, kinases, TF's such as c-fos & c-jun). The proto-oncogene may also include inhibitors of apoptosis (eg. bcl2) such that the mutant cell can avoid regulated cell death.

A single recombination event to produce an oncogene is sufficient to induce a tumor since oncogenes are dominant mutants.

18. Throughout this course we have studied the "Central Dogma" of molecular biology, "DNA -> RNA -> Protein". However, molecular biologists now believe that the history of life began with RNA (or something like it) which then 'invented' proteins and later used proteins to 'invent' DNA. Give one specific example for each of the following lines of evidence for an ancient 'RNA World':

a. (2pts) Catalytically active RNA molecule that can function like an 'enzyme'.

<u>Specific RNA:</u>	<u>Reaction catalyzed:</u>
a. Spliceosomal snRNPS (U1, etc) b. large rRNA c. RNase P	a. splicing exons/removal of introns b. peptide bond synthesis (peptidyl transferase) c. trimming of tRNA

b. (2pts) 'Activation' of an amino acid by a ribonucleotide for peptide bond synthesis

<u>'Activating' ribonucleotide:</u>	<u>Enzyme(s) responsible for activation of amino acids:</u>
ATP or tRNA	Aminoacyl-tRNA synthetases

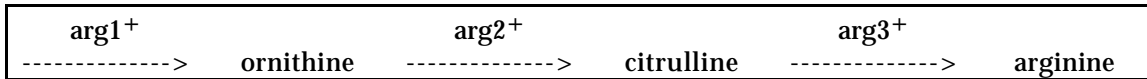
c. (2pts) Chemical modification of a nucleotide to make DNA instead of RNA.

<u>Chemical modification:</u>	<u>Effect on DNA stability:</u>
a. U Methylation -> T b. ribo -> 2'-deoxyribo	a. increase stability of sequence = Allows repair of U's resulting from deamination of C b. increase stability of polymer = reduces rate of hydrolysis of S-P backbone due to catalysis by 2'OH

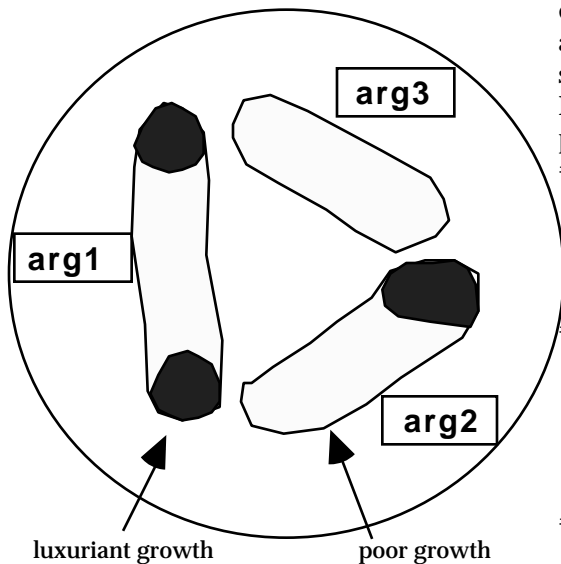
19. (7 pts) The following diagram shows a *Drosophila* polytene chromosome, and the bars below it show the region of DNA deleted in deletions 1 through 6. Recessive mutations t, u, v, w, x, y, and z are known to map to this portion of the chromosome, but their exact order is not known. The table shows complementation data for each deletion with each mutation ("-" means the mutant phenotype is observed, and "+" means the wild-type phenotype is observed). Use these data to infer the position of each mutation, and write its letter in the appropriate box above the polytene chromosome.

	t	z	v	u	x	y	w
Del 1	-	+	+	+	+	+	-
Del 2	-	-	-	+	+	+	-
Del 3	+	+	-	+	+	+	-
Del 4	+	-	+	+	-	+	+
Del 5	+	+	+	-	-	-	+
Del 6	+	+	+	+	-	-	+

20. A haploid strain of *Neurospora* carries a new auxotrophic mutation in the arginine biosynthetic pathway. It grows on medium supplement with citrulline and arginine, but not ornithine.



- a. (2 pts) Which gene is mutated in this strain? **arg2**
- b. (2 pts) Mutations in which genes will complement this new mutation? **arg1 and arg 3**
- c. (3 pts) In the following cross-feeding experiment, 3 different arg mutant strains are streaked on a plate containing a small amount of arginine. Write in each box the name of the gene mutated in each strain:



- d. (6 pts) 3 prototrophic revertants were isolated from an arg auxotrophic mutant strain. Each is crossed with a standard wild-type strain and 1000 progeny examined. Explain the origin of each revertant, given the following progeny data:
- #1: 250 arg auxotrophs, 750 prototrophs **unlinked suppressor segregating independently of the original arg mutation: 1/2 of the progeny will be recombinant and 1/2 of those will be the arg auxotrophic mutation without the suppressor**
- #2: 100 arg auxotrophs, 900 prototrophs **linked suppressor, 20 map units from the original arg mutation. The auxotrophic class has recombined the arg mutation away from the suppressor. The reciprocal cross-over is not detected, so the RF is twice the frequency of auxotrophs.**
- #3: 0 arg auxotrophs, 1000 prototrophs **reversion of the original mutation. Since no auxotrophic progeny are detected, the reversion event is very tightly linked to the original mutation, perhaps at the site of the original mutation itself.**