PLNT3140 INTRODUCTORY CYTOGENETICS
FINAL EXAMINATION

December 13, 2014
Time: 18:00 - 20:00
Location: Engineering E2-165

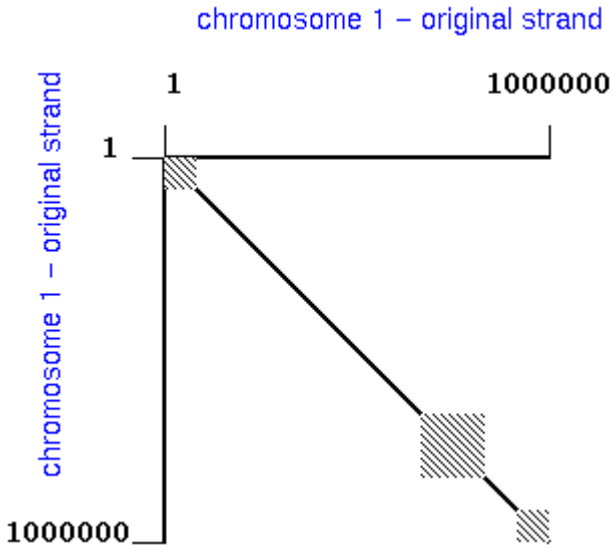
Answer any combination of questions totaling to exactly 100 points. If you answer questions totaling more than 100 points, answers will be discarded at random until the total points equal 100. There are 10 questions to choose from, totaling 120 points. This exam is worth 35% of the final grade.

Ways to write a readable and concise answer:
i. Just answer the question. Save time by specifically addressing what is asked. Don't give irrelevant background if it doesn't contribute to the question that was asked.
ii. Avoid stream of consciousness. Plan your answer by organizing your key points, and then write a concise, coherent answer. Make your point once, clearly, rather than repeating the same thing several times with no new information.
iii. Point form, diagrams, tables, bar graphs, figures are welcome. Often they get the point across more clearly than a long paragraph.
iv. Your writing must be legible. If I can't read it, I can't give you any credit.

---

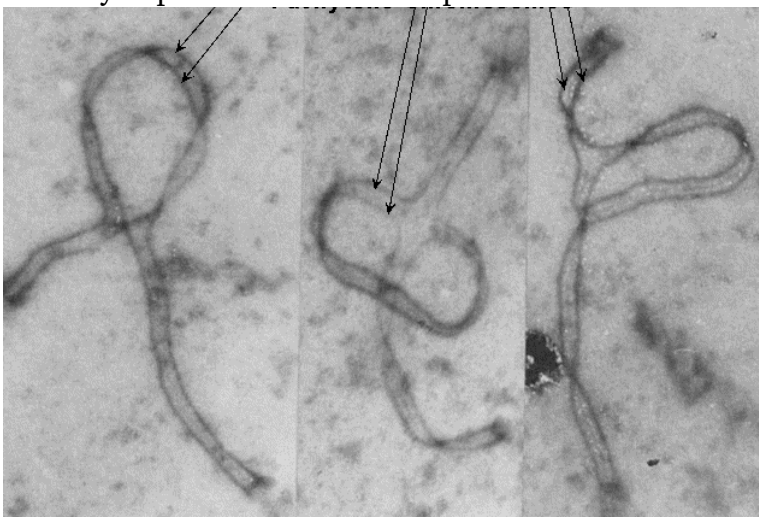
1. (10 points) The structure and evolutionary history of entire chromosomes can be studied using dot-matrix programs such as DXHOM. For example, comparison of an entire chromosome with itself might give a plot like that shown at right.

Explain why we see these "diamond" arrays of parallel diagonals.



2. (10 points) How are doubled haploid plants produced in the lab? What advantages do doubled haploid plants offer to geneticists and plant breeders?

3. (10 points) The chromosomes below are from cells at pachytene.
a) Why do the chromosomes appear as two threadlike-structures, rather than four?
b) What is the most likely explanation for the loops seen in each?



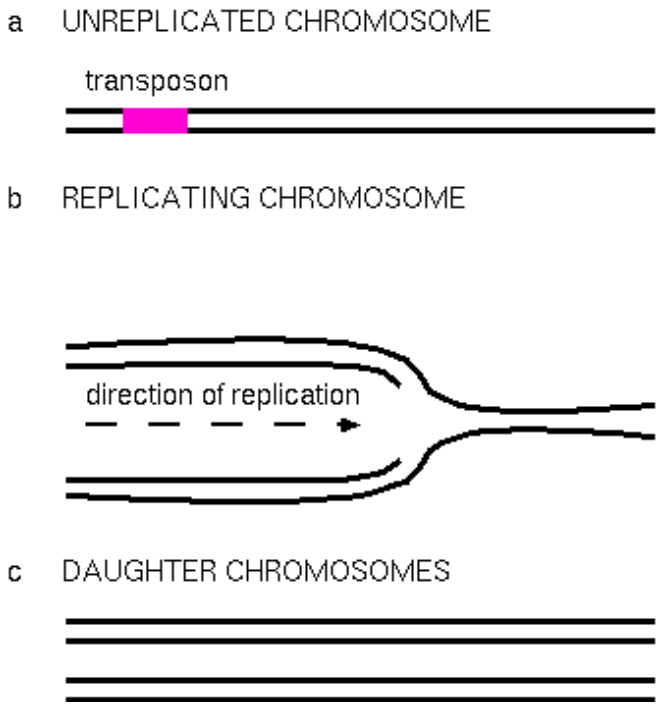
4. (15 points) Most molecular markers used for mapping in eukaryotes are selectively neutral. Cite two independent reasons why this is true. Also, explain why this matters in genetic mapping.

5. (10 points) Bacterial artificial chromosomes (BAC) are plasmids designed to facilitate the cloning and manipulation of _____a_____. A BAC vector will typically have cloning sites for enzymes such as Not I (5'GC^GGCCGC3'), which is expected to cut once every _____b_____ base pairs. The cloning site in the vector usually contains a _____c_____ that interrupts a selectable marker. For example, some BAC vectors contain a gene such as SacB, whose expression is toxic to bacterial cells grown on sucrose. When used for cloning, the SacB gene is removed by a restriction digest. Thus, one can select for clones containing inserts by _____d_____. One advantage of BACs, compared to linear YACs, is that BACs are circular, which are less susceptible to _____e_____ than linear chromosomes would be.

6. (5 points) When designing a microarray, the goal is to create 60-mer oligonucleotides, each of which recognizes a specific gene. Given that a gene may consist of coding sequences, introns, and 5' and 3' non-coding regions, which regions would be the best choice as the source of the 60-mer sequence for a gene? Explain your reasoning.

7. (5 points) Stable triploids are not found in sexually-reproducing species, but are found in parthenogens. Explain why.

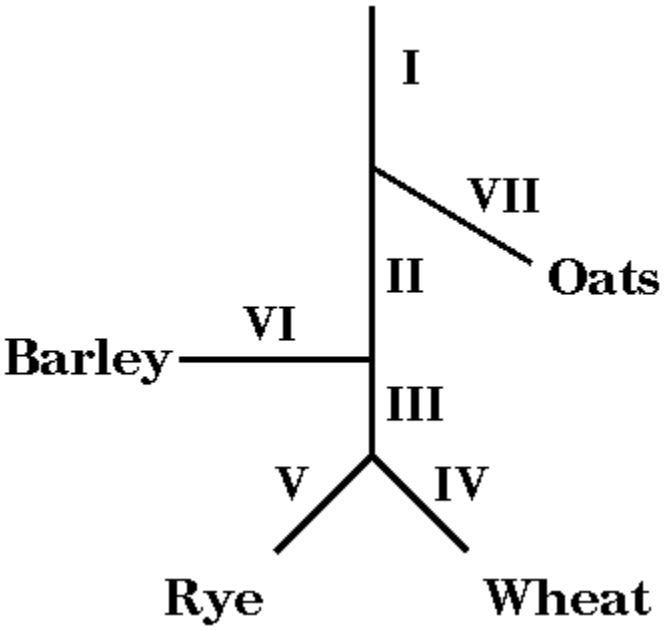
8. (5 points) Redraw the picture below, showing how transposition of a transposon during DNA replication can result in a net increase in the number of transposons.



9. (10 points) Explain how changes in chromosomal structure, such as duplications, deletions, translocations and inversions, help drive the process of speciation. You do not need to go into detail on defining what these changes are eg. don't bother drawing diagrams of meiotic products resulting from translocations. Rather, concentrate on the evolutionary consequences of these changes.

10. (10 points) You are beginning a new project studying ribosomal RNA genes, which are typically present in > 200 copies per haploid genome. The project will involve a lot of Southern blots, which normally require a 20 hour hybridization time to detect a single copy gene. You realize that it should be possible to do your Southern blots in a shorter time, because of the fact that rRNA genes are present in high copy numbers. If you want to get bands with the same intensity as you would get for a single copy gene, how long should you hybridize? Hint: Consider the definition of C_0t.

11. (15 points) In the experiments of Flavell and co-workers, C_0t analysis was employed to compare the relative amounts of interspersed repetitive DNA families in four cereal species. As we discussed, the authors defined seven distinct classes of repetitive sequence families, I - VII. For example, Class I refers to repetitive sequences found in the common ancestor of all 4 species, while Class VII refers to repetitive sequences that arose after the divergence of Oat from its common ancestor with Barley, Wheat and Rye. They performed C_0t curves to determine, for example, the percentage of Class II sequences present in Rye, Wheat and Barley, or the percentage of Class III sequences found in Rye or Wheat.



In principle, you obtain the same information using 60-mer microarrays. A critical consideration in this case is that while a substantial percentage of the wheat and barley genomes have been sequenced, there is relatively little sequence available from rye and oat genomes, at this writing. Consequently, it should be possible to construct microarrays containing 60-mers representative of a substantial percentage of the Wheat and Barley genomes.

a) In your exam booklet, rewrite the table below, filling in roman numerals of any class of sequences that would show up as repetitive using either the Wheat or Barley microarrays, when hybridized with labeled genomic DNA from Wheat, Rye, Barley or Oat.

	microarray:	Wheat	Barley
labeled-DNA:			
Wheat			
Rye			
Barley			
Oat			

b) What additional things would you learn from microarrays that you wouldn't learn from C_0t analysis?

12. (5 points) When chromosome painting was developed, only five fluorescent dyes were available that would fluoresce at distinct wavelengths, such that their presence could be measured simultaneously in the same in-situ hybridization experiment. However, there are 24 unique human chromosomes. How did the inventors of chromosome painting get around the problem of uniquely detecting each chromosome?

13. (10 points)

a) Draw a diagram illustrating how chromosomes pair during meiosis in cells heterozygous for a reciprocal translocation. Feel free to use colors, shading or hatching to represent homologous chromosomes.

b) Draw a diagram illustrating the orientation of reciprocal translocation products from a), at metaphase, assuming an Alternate configuration.