39.314 INTRODUCTORY CYTOGENETICS

MID-TERM EXAMINATION

1 p.m. to 2:20 p.m. Tuesday, October 21, 2003

This examination is worth 15% of the course grade. There are 9 questions totalling 100 points.

Hand in these question sheets along with your exam book.

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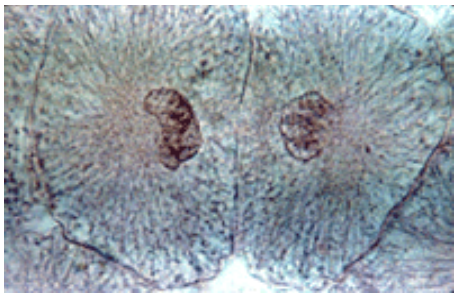
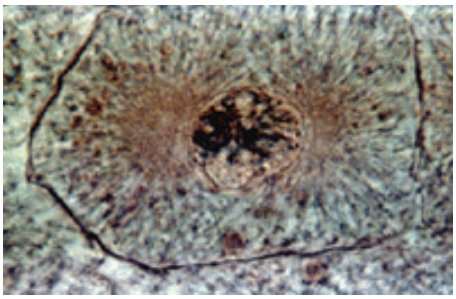
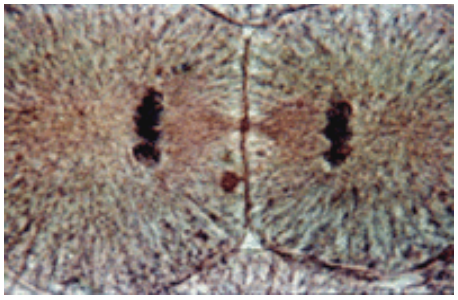
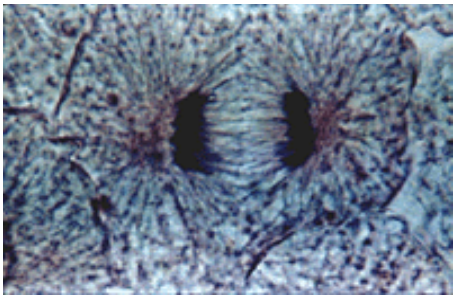
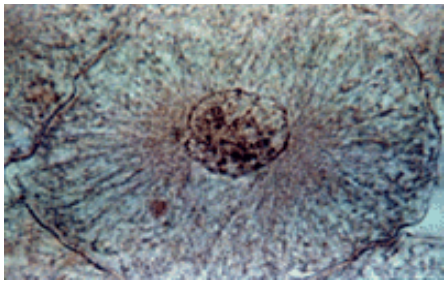
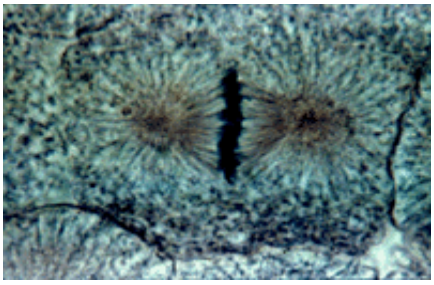
1. (12 points) Identify each of the stages of mitosis depicted below. (Hint: Interphase is depicted both prior to and after a cycle of mitosis.)

A 	***B*** 
C 	D 
E 	F 

2. (20 points) Definitions:

a) centrosome
b) telomere
c) nucleosome
d) germ-line cell
e) chromatin domain

3. (8 points) Complete the sentences below:

The main steps for studying chromosomes are:

a) Collection of material to _____
b) Pretreatment to _____
c) Fixation to _____
d) Staining to _____
e) Slide preparation for optimal viewing
f) Data recording and micrometry

4. (5 points) Why is there a potential conflict between the process of cell division and the process of gene expression?

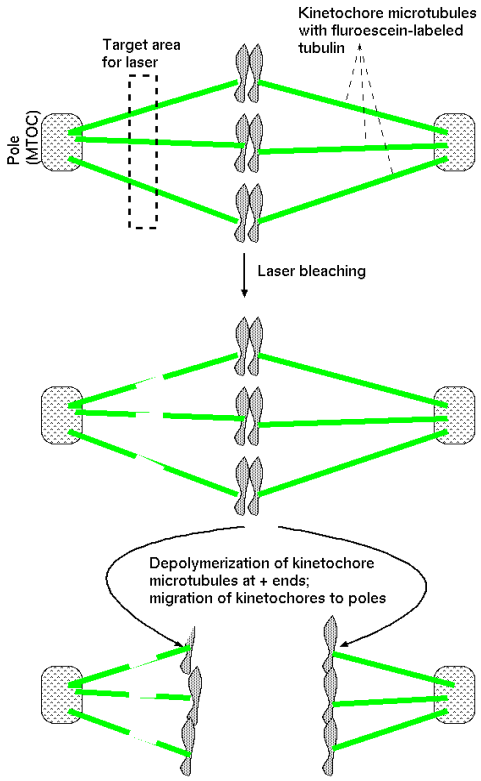
5. (5 points) Replication origins tend to be rich in _____ , which makes it easier for DNA double helices to open up, leading to the initiation of replication.

6. (10 points) The FACT ("FAcilitates Chromatin Transcription") protein consists of two polypeptides Spt16 and SSRP1. Both polypeptides are highly conserved in eukaryotes. Experiments have revealed the following:

a) FACT binds nucleosomes
b) Spt16 has a highly acidic (ie. negatively-charged) N-terminus
c) FACT can bind a H2A-H2B dimer *in-vitro*. It does not bind H3-H4 dimers.
d) Nucleosomes incubated with FACT have only about half as much H2A-H2B as H3-H4.
e) FACT is required for RNA transcription, DNA replication and DNA repair.

Propose a model for the activity of FACT that accounts for all five observations.

7. (10 points) State two alternative hypotheses that could be distinguished by this experiment. What prediction is made by each hypothesis, and which is the correct hypothesis?



8. (10 points) During attack by pathogens, plant cells actively transcribe a battery of defense genes whose products are needed for limiting the growth of fungi or bacteria, strengthening cell walls, degrading toxic compounds, or containing the spread of pathogens. Klaus Hahlbrock and coworkers have found that when parsley cells are inoculated with spores from the fungus *Phytophthora megasperma*, genes required for mitosis (eg. histone genes, cell-cycle regulators) actually showed <u>decreased</u> transcription (<u>Plant J</u>.8:865-976). One could argue that this shutdown of mitosis in response to pathogenic attack is to conserve limited energy needed to fight the fungus, but there is another, more fundamental reason. In light of what we know about chromatin, how would entering mitosis compromise the cell's ability to respond to pathogenic attack? (Be brief).

9. (20 points)

a) Draw a diagram illustrating the process of DNA replication. Make sure that your diagram shows <u>two</u> adjacent replicons (ie. 1 replicon = 1 origin of replication flanked by two replication forks). Label leading and lagging strands.

b) The figure below shows the result of pulse-labeling experiments with the fluorescent nucleotide analog bromodeoxyuridine (BUdr). Examples of normal chromosomes (ie. that have not undergone sister chromatid exchange) are pointed to with arrows. Referring to your diagram from part **a**, explain how these results support the model that the eukaryotic chromosome is a single linear DNA molecule. Put another way, how does your model of DNA replication lead to the partitioning of all label on one chromatid, and no label to the other chromatid, even when there are dozens or even hundreds or replication origins on each chromosome?

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            1 cell                fresh media   1 cell
      +BUdr  cycle                 -BUdr         cycle
cells ----->------>labeled cells ------------>-------->
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