PLNT3140 INTRODUCTORY CYTOGENETICS

MID-TERM EXAMINATION

1 p.m. to 2:20 p.m.   Tuesday, October 24, 2006

This examination is worth 100 points, for 15% of the course grade.

Hand in these question sheets along with your exam book.

1. (10 points) Draw a diagram of the eukaryotic cell cycle, and use a few keywords or phrases to indicate the events that characterize each stage. Also, indicate which stage or stages correspond to interphase.

2. (20 points) During prophase, _____i_____ migrate to opposite poles of the cell, and extend microtubules throughout the cell. _____ii_____ also occurs during prophase. At this point, a spindle fiber may attach to a _____iii_____, which stabilizes the spindle. Congression of chromosomes to a single plane is the result of _____iv_____ . _____v_____ begins when chromosomes separate, and progress to the opposite poles. The energy for movement of the chromosomes comes from depolymerization of _____vi_____ which make up the spindle fibers. During telophase, the _____vii_____ reforms such that each chromosome is packaged as a separate vesicle. The division of the cytoplasm, referred to as _____viii_____, also occurs in telophase. Next, chromosomal vesicals merge to form a complete nucleus. Chromosomes _____xi_____, but because of the compartmentalization of the nucleus chromosomes remain in well-defined _____x_____.

3. (10 points) Cite two reasons why is synapsis essential to meiosis.

4. (5 points) Why does heterochromatin stain more darkly than euchromatin?

5. (10 points) Histones are among the most highly conserved proteins in evolution. For example, Histone H4 is 102 amino acids long, with only two amino acid substitutions found between pea and cow. Other chromatin-associated proteins have numerous mutations, when proteins are comared between distantly-related species. Suggest an explaination for these observations.

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 <u>not </u>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) Prokaryotic genomes could be thought of as one big domain, whereas eukaryotic genomes are divided into many hundreds or thousands of domains. At the same time, prokaryotes are generally single-celled, whereas eukaryotes have evolved into multicellular organisms with highly-differentiated tissues. What would be a possible connection between these two observations?
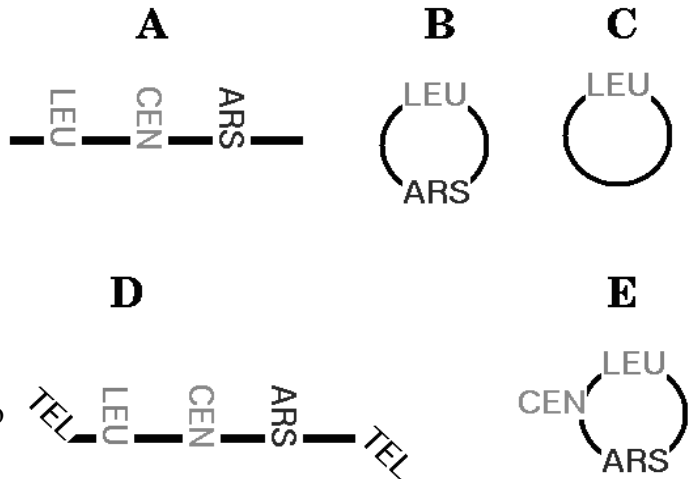

8. (10 points) The sequence below shows the Simian Virus 40 origin of replication, as annotated in Sumitra et al. (1986) <u>Mol. Cell. Biol</u>. 6:1663-1670. Recalling what you know about DNA, is there a feature of this sequence which might be particularly important to facilitating formation of a replication fork? Explain your answer.

```
           Early Palindrome     T antigen binding                  AT
           ------>  <-----     ----> ----> <---- <----    ----------------
GGCCTCCAAAAAAGCCTCCTCACTACTTCTGGAATAGCTCAGAGGCCGAGGCGGCCTCGGCCTCTGCATAAATAAAAAAAATTA
CCGGAGGTTTTTTCGGAGGAGTGATGAAGACCTTATCGAGTCTCCGGCTCCGCCGGAGCCGGAGACGTATTTATTTTTTTTAAT
```

9. (15 points) The figure at right shows several recombinant DNA constructs containing sequences including the yeast Leucine synthase gene (LEU), the yeast centromere (CEN), the yeast origin of replication (ARS) and the yeast telomere (TEL). The bottom figure illustrates several experiments in which yeast mutants deficient in leucine biosynthesis (ie. *leu* ⁻) were transformed with one of these constructs and plated first on complete media, and later plated on minimal media to test for growth without leucine. For each of the five constructs (A - E), indicate which of the experimental results (1-4) would be seen and explain why.

A)

B)

C)

D)

E)

A **B** **C**

D **E**

1)

many generations

complete media

minimal media

2)

many generations

complete media

minimal media

3)

many generations

complete media

minimal media

4)

many generations

complete media

1 generation complete media

minimal media