Biology 200 Section
Midterm Exam – October 20, 2010
Total time: 75 min. Worth a total of 44 marks
22% of Course Grade

If there are any marking mistakes in your exam, your instructor can remark it. If you want that your exam remarked:
1. Please compare your exam to the answer key posted on the VISTA site.
2. Please put in writing your request, attach it to the exam and give it to your instructor.
3. Leave the exam (with writing request) with your instructor during class, or office hours.
4. Your instructor will remark the whole exam, so marks can go up or marks can go down!!
Please hand-in all exams with requests to your instructor by **November 12th, 2010**.

Except for Q 6, answer all questions on this exam paper. Use black or blue PEN only.

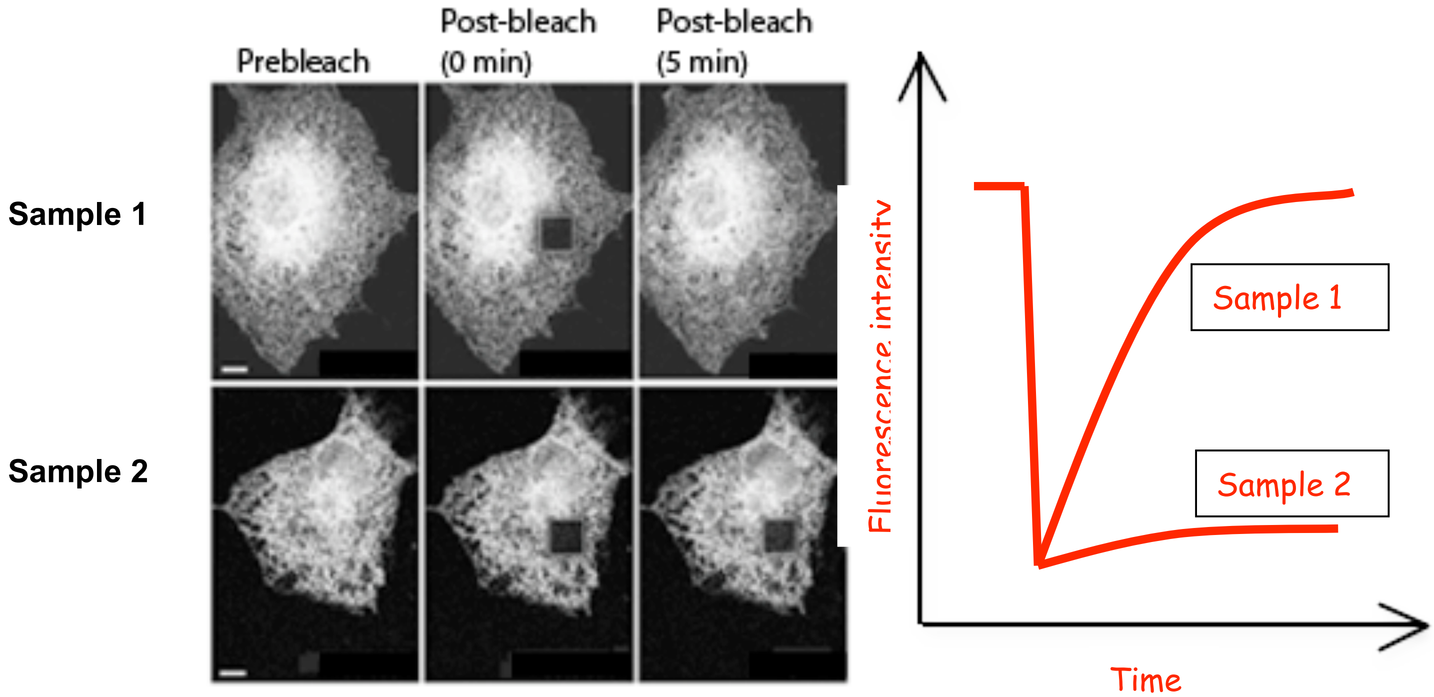
Question	1	2	3	4	5	6	TOTAL
Student Score							
Maximum Score	5	7	6	6	5	15	44

Question 1 (5 marks)
Match the experiment with the type of microscopy that you could use to perform that experiment.

Experiment	Answer	Type of microscopy
See viruses crossing nuclear pore complexes	A	A. Transmission electron microscopy
Localize a green fluorescent protein-tagged plasma membrane protein.	D	B. Scanning electron microscopy
Visualize the surface of a fungal cell.	B	C. Bright field light microscopy
Count the number of chromosomes in a species.	C* (B or D)	D. Confocal (fluorescence) microscopy
Visualize the distribution of ribosomes on the endoplasmic reticulum	A	

*SEM or confocal are also acceptable

Question 2 (7 marks)



The images above show mammalian cells expressing a fluorescently-tagged protein in the endoplasmic reticulum. A square region was photobleached and the fluorescence recovery after photobleaching (FRAP) was recorded.

a. On the graph above, label the axes and draw the recovery curves for samples 1 & 2 shown in the images. (2 marks)
   1 mark – label axis
   2 marks – for each curve (label the curves)

b. Explain what FRAP data tells you about the protein's environment in each cell and propose an explanation for the difference in recovery of fluorescence between the two samples. (4 marks)

FRAP demonstrates the mobility of the fluorescent protein in the fluid bilayer. (2 marks)
Possible explanations:

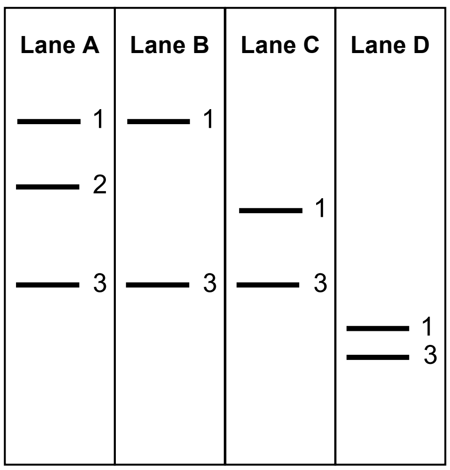
The protein in sample 2 is anchored to another protein. (2 marks)

<u>Other possible explanations</u>:
- Fluidity of membrane without explanation (1 mark)

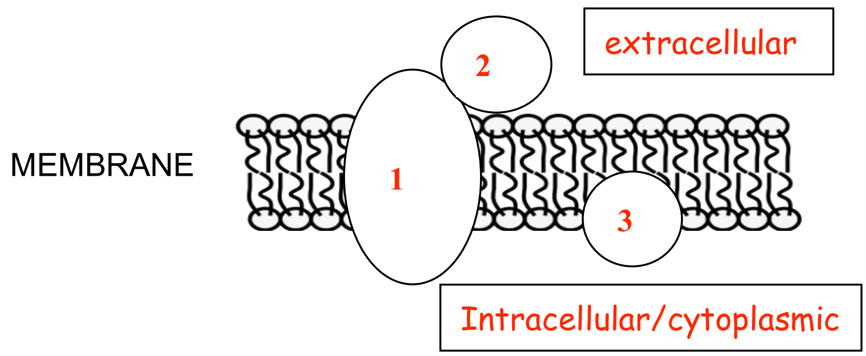
OR
- Fluidity / mobility of protein within the lipid bilayer, unsaturated vs saturated fatty acid tails, longer vs shorter fatty acid tails, cholesterol, etc. (2 marks)

Question 3 (6 marks)
Shown below are drawings representing the results of the gel electrophoresis analysis of three membrane proteins extracted from cells treated in a variety of ways:

Lane A: intact cells with no treatment
Lane B: intact cells after treatment with a solution of high salt concentration
Lane C: intact cells treated with trypsin.
Lane D: permeabilized (broken) cells treated with trypsin.



Based on these results, draw <u>and label</u> the arrangement of the proteins on the membrane on the diagram shown below, <u>and indicate</u> the type (integral or peripheral) of membrane protein:



- 1 mark for labelling "extracellular/intracellular"
- 2 marks for each protein type:
    - 1=integral/transmembrane;
    - 2=peripheral (i.e. associated with another protein)
    - 3 = integral/monolayer
- 3 marks for arrangement/correct drawing (1 mark each)

Question 4 (6 marks) – write in the space provided.

For each of the experimental observations below, please explain what it tells us about nuclear structure/ function. Why?

A. When a 30nm fiber is treated with a high salt solution, an 11nm, beads-on-a-string fiber is observed in TEM.

- Chromatin is packed as beads on a string, i.e. DNA 'string' is wrapped around nucleosomes (1 mark).

- Salt treatment removes histone H1 from 30 nm chromatin and reveals nucleosomes (1 mark).

  <u>Note</u>: [½ mark for mentioning histone H1, full mark for mentioning that salt removes histone H1. 1 mark for a statement like "some factor, most likely a protein, can be removed from the 30nm by high salt to give the 11nm"]

B. Treating a batch of mammalian cells with hormones results in drastic increases in gene expression. TEM micrographs of these cells show increases in the ratio of euchromatin to heterochromatin in the nucleus.

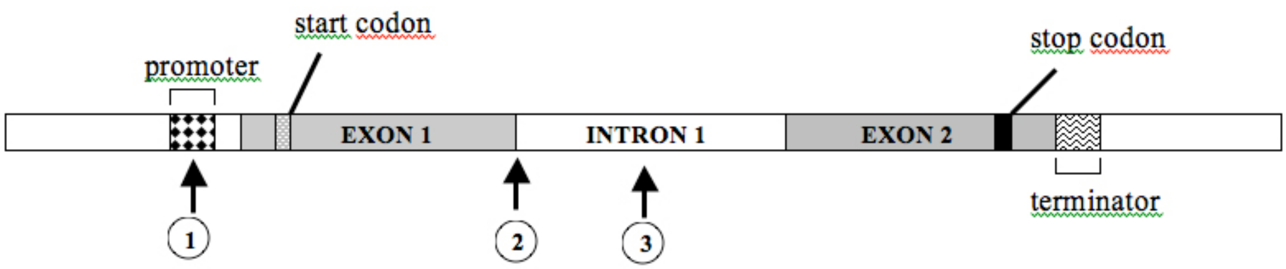
- The change in the ratio of euchromatin : heterochromatin is correlated with changes in transcriptional activity. (1 mark).
  [½ mark if you said "an increase in the amount of euchromatin causes an increase in gene expression", since only correlation, not causation is shown.]

- This suggests that euchromatin is the transcriptionally active form of chromatin. (1 mark).

C. Nucleoli become heavily radiolabeled when radioactive ribonucleotides are provided to the cell.

Radioactive ribonucleotides move into the nucleus (0.5 marks), are incorporated to rRNA (1 mark) as it is being transcribed in the nucleolus [ribosomal subunits assembly in nucleolus] (0.5 mark).

Question 5 (5 marks)
Shown below is a diagram of a typical eukaryotic gene, which codes for protein X.



What would be the effect on the synthesis of the protein and its structure if:

a) A mutation occurs in the region labeled 1. (1 mark).
The promoter would be disrupted and the gene would not be transcribed so no protein would be made. [½ for just saying gene won't be transcribed, not mentioning protein]

b) One base pair is deleted in the region labeled 2. (2 marks).
The intron/exon junction sequence could be disrupted and the intron could be retained. The protein will be produced but will have a new component several amino acids long in the middle. This will interfere with the folding of the original protein, thus the structure of the protein would be modified. (2 marks)

[- ½ for not mentioning structure of the protein, e.g. "a very different protein would be made"
-1 for not mentioning protein at all]

Other acceptable answer: The protein produced might be truncated if a STOP codon is encountered within the intron that didn't get removed, thus a new protein with a very different structure.
or
This leads to mRNA degradation so no protein is made or this leads to retained, potentially out of frame, sequence being added to the protein.

c) One base pair is introduced in the region labeled 3. (2 marks).
Addition of a base pair should cause no change to the final protein as the intron is removed anyway (1mark). Here, Protein X would be produced normally and there would not be any effect on its structure. (1mark)
Optional acceptable answer: IF the added base is in the sequence required for the formation of the lariat, the intron would not be removed (1mark). Here, the new protein with additional amino acids in middle, could have different folding and hence different structure (1mark) or the new protein might be highly truncated if a STOP codon is encountered within the retained intron (1mark). [-1 for not mentioning protein].

Question 6. Essay 15 marks
Compare and contrast the transport of molecules across the plasma membrane of a typical animal cell with the transport of molecules across the nuclear envelope.
Organization - 5 marks
- Thesis statement- 1 mark
- Organization of ideas into paragraphs, introduction and conclusion - 2 mark
- Quality of writing, including clarity and correct use of terminology- 2 marks

Content - 10 marks
What is the quality of the scientific argument presented? Are the main points well supported with specific information?
2 marks – contrast
2 marks – comparison
3 marks – NPC detail
3 marks – plasma membrane detail

Possible Thesis Statements:
1. The mechanisms of transport of molecules across the plasma membrane or the nuclear envelope both represent controlled movement across cellular barriers, however there are fundamental differences reflecting the structure and functions of these barriers.
2. The transport of molecules across the plasma membrane, and across the nuclear pore complex have essential similarities because integral membrane proteins and protein complexes are required, however different mechanisms are involved to perform the functions of these membranes.

Similarities:
- Molecules move across membranes and across the nuclear pore complex in both directions.
- Small molecules can pass both the PM and NPC by diffusion.

- Larger molecules need "help" to go through either the PM or the NPC, and they will be actively transported across by integral membrane proteins.
- Some larger &/or charged molecules will be actively transported through both the PM and the NPC.
- The NPC and the PM both contain proteins.
- Transport in both the PM and NPC may or may not require energy.

Differences:

- Lipid bilayers are impermeable to solutes and ions, and so the PM needs proteins to be able to transport them. These membrane proteins can be transporters or channels. They allow molecules to cross the PM by either passive or active transport.
- For molecules to cross the nuclear envelope, they need to cross the NPC. Morphologically, the NPC model is composed of a large number of protein subunits, as well as protein fibrils found in the cytosolic side of the nuclear envelope. Fibrils are not present in channels found within the membrane.
- The fibrils in the NPC are involved in active transport of molecules, whereas these fibrils are not found in the PM.
- Active transport in the PM requires transporter proteins. These proteins are phosphorylated/dephosphorylated to be able to move molecules actively across the PM. These proteins go through a conformational change that mediates the transport of molecules across membranes.
- Active transport in the NPC is different than the one in the plasma membrane, as it involves the opening of the nuclear pore to move molecules across the nuclear envelope.
- Plasma membrane channels are smaller in diameter than the NPC opening.
- Molecules are able to cross the PM by diffusion, facilitated diffusion, or active transport. To be able to cross the NPC, small molecules can also cross by diffusion, but larger molecules are imported into the nucleus by active transport.
- Some molecules (i.e. large proteins) need to have an NLS (nuclear localization signal) to get into the nucleus. This signal is not required for molecules to cross the plasma membrane.
- Energy source: ATP in plasma membrane vs. GTP in NPC