Biology 200
Sample Midterm Exam #2 – Peer Tutor Review Midterm
Total time: 75 min.

Section: _____

Family Name: _____ Given Name: _____Student #: _____
(please print)

Instructions:

1. Use black or blue PEN only. Exams that are written in pencil, or that have corrective tape on them will not be eligible for re-grading.
2. There are 6 questions on 9 pages. Answer all questions in this exam booklet.
3. You are allowed an 8.5x11 inch, double-sided, hand-written memory aid for this exam. All memory aids that do not conform to these rules will be taken away.
4. You will have 75 minutes for this exam.
5. Please write your name and student number on the **first AND last page of this exam**. Your grade will be written on the last page.

Codon table for your reference:

GCA GCC GCG GCU	AGA AGG CGA CGC CGG CGU	GAC GAU	AAC AAU	UGC UGU	GAA GAG	CAA CAG	GGA GGC GGG GGU	CAC CAU	AUA AUC AUU	UUA UUG CUA CUC CUG CUU	AAA AAG	AUG	UUC UUU	CCA CCC CCG CCU	AGC AGU UCA UCC UCG UCU	ACA ACC ACG ACU	UGG	UAC UAU	GUA GUC GUG GUU	UAA UAG UGA
Ala	Arg	Asp	Asn	Cys	Glu	Gln	Gly	His	Ile	Leu	Lys	Met	Phe	Pro	Ser	Thr	Trp	Tyr	Val	stop
A	R	D	N	C	E	Q	G	H	I	L	K	M	F	P	S	T	W	Y	V	

Figure 7-24 Essential Cell Biology 3/e (© Garland Science 2010)

Question 1 (5 marks)

Classify each of the images shown into the table below, based on the type of microscopy and the organelles that you can SEE. Be aware that each image may fit into more than one category and not all categories may be present. Marks will be deducted for wrong answers.

Image 1:

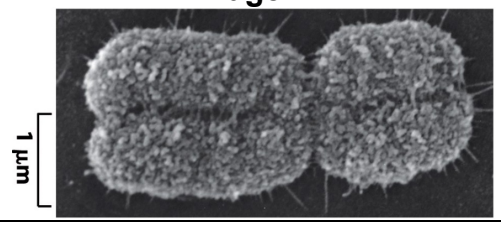


Image 2:

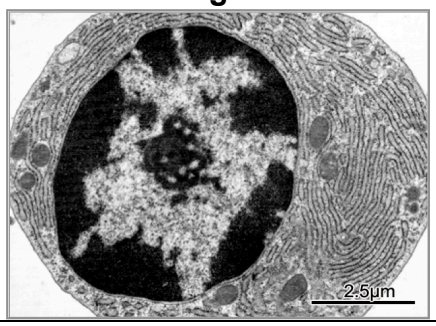


Image 3:

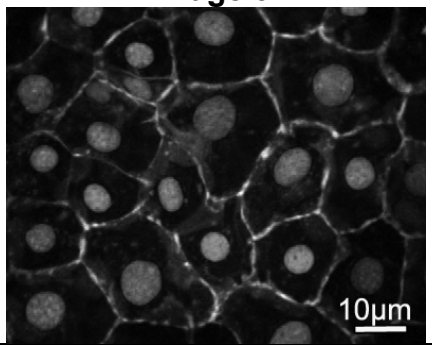
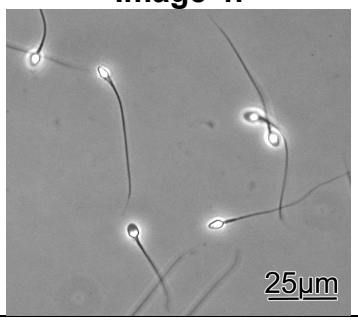


Image 4:



Type of microscopy:	
Brightfield light microscopy	**4**
Fluorescence microscopy	**3**
Transmission electron microscopy	**2**
Scanning electron microscopy	**1**

Organelles:	
Plasma membrane	**4, 3, 2**
Nucleus	**3, 2**
Nucleolus	**2**
Endoplasmic reticulum	**2**
Mitochondria	**2**
Cytosol	**2,**
Chloroplast	
Heterochromatin	**2, (3)**
Mitotic Chromosome	**1**
Flagella	**4**

Note: There may be more organelles visible than listed here.

Question 2 (6 marks)

For each of the experimental observations below, please state what it tells us about cellular structure and/or function, and explain why.

A. 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).

B. When enzymes isolated from cells are heated, they can no longer catalyze a reaction.
Applying heat denatures isolated proteins by disrupting all the non-covalent bonds that maintain the secondary and tertiary structures. This means that 3D protein structure is important for substrate binding/enzyme function.

C. If the NLS from a nuclear protein is experimentally added to hexokinase (a cytoplasmic protein), the altered hexokinase is detected in the nucleus.
This means that the NLS sequence of a nuclear protein is sufficient to drive nuclear import because adding it alone to a normally cytoplasmic protein caused it to localize in the nucleus.

D. When a mutation is introduced at an exon-intron junction of the b-globin gene, this results in the production of a longer mature mRNA transcript.
This means that the sequence at the exon intron junction is critical for recognition by the spiceosome to remove the intron of the pre-mRNA. The mature mRNA is longer because it contains the intron sequence that is normally spliced out.

E. If mammalian cells are treated with hormones and then imaged using TEM, the results show increases in the amount of euchromatin relative to heterochromatin in the nucleus.
The increased euchromatin compared to heterochromatin correlates with increases in transcriptional activity in response to hormone treatment. This suggests that hormone treatment stimulates transcription and euchromatin is the transcriptionally active form of chromatin.

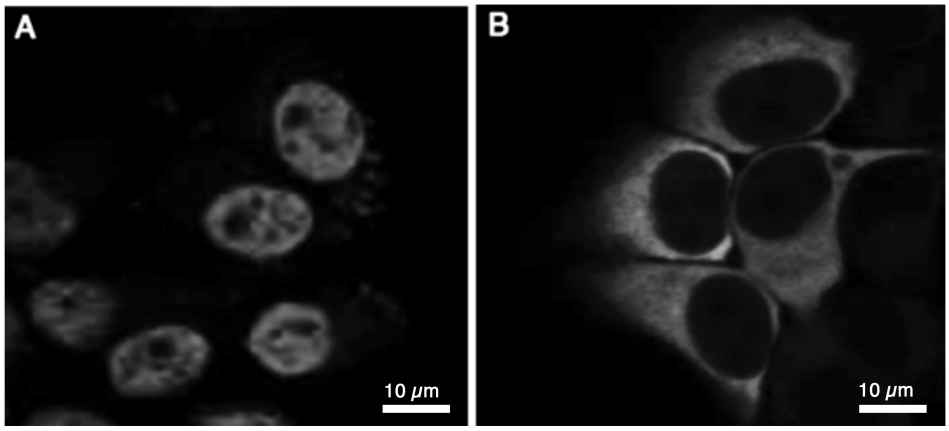
F. If cells are treated with SDS (a strong detergent) membrane proteins can be separated form phospholipids.
SDS is structurally amphipathic and can form detergent micelles. Detergent miscelles can separate membrane proteins from membrane phospholipids due to their long hydrophobic tails which associate with the hydrophobic regions of membrane proteins and the fatty acid tails of phospholipids which causes the membrane to disassemble.

Question 3 (6 marks)

A novel nuclear protein has a molecular weight of 100 kDa. Below in Figure A is an image showing the normal localization of this protein in mammalian cells. A mutation in the DNA of this protein changes its cellular localization as shown in Figure B. When the DNA sequences of the wild-type (normal cells) and mutant cells were compared there was only one difference in the sequences: only the underlined nucleotide below was different.

Beginning of the coding region of the template strand of normal cells:
3'T A C C A G T **C** T T C C…….5'

Beginning of the coding region of the template strand of mutant cells:
3'T A C C A G T **G** T T C C…….5'



A. Does the mutation introduce changes in the primary structure of this protein? Explain. (2 marks)

Yes, the mutation will change the mRNA codon AGA in the normal cell for ACA in the mutant cell, which will result in a change in amino acid Arg for Thr.

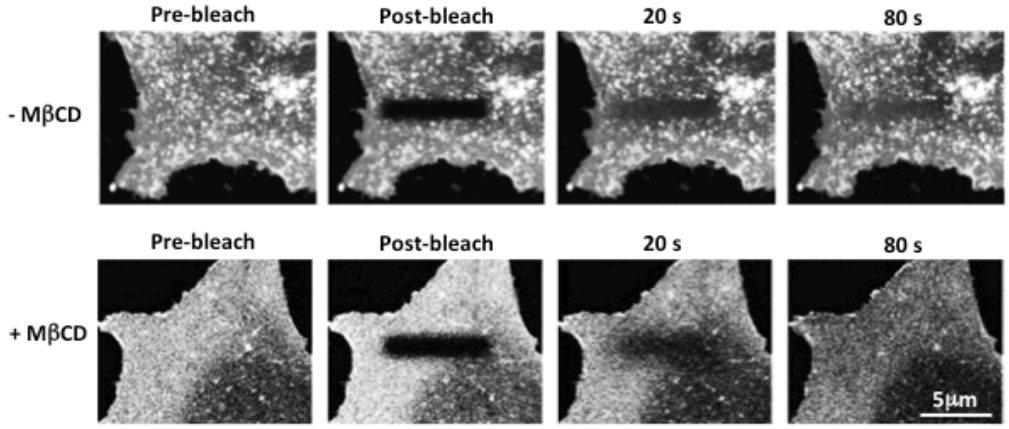
B. Describe what the data in Figure A and B above shows. (1 mark)

The image in Figure A shows that the fluorescence localizes in the nucleus.
The image in Figure B shows that the fluorescence localizes in the cytoplasm.

C. Explain why this mutation can change the cellular localization of this protein. (3 marks)

The mutation resulted in changing the amino acid Arg (positively charged) to Thr (uncharged polar) (1 mark). The amino acid Arg in the normal protein might have been part of an NLS (1 mark), which is necessary for nuclear import and consist of a short sequence of positively charged amino acids. A mutation of a positive charged amino acid to an uncharged polar will make the NLS non functional for nuclear import.

Question 4 (7 marks)
RasGTPase (Ras) is a lipid-linked, plasma membrane-associated protein involved in cell signalling. Goodwin et al. tagged Ras with GFP and expressed it in cells to examine its mobility by Fluorescence Recovery After Photobleaching (FRAP). They compared the mobility of Ras in the presence (+) or absence (-) of a drug called methyl-β-cyclodextrin (MβCD) that removes cholesterol from the membrane.



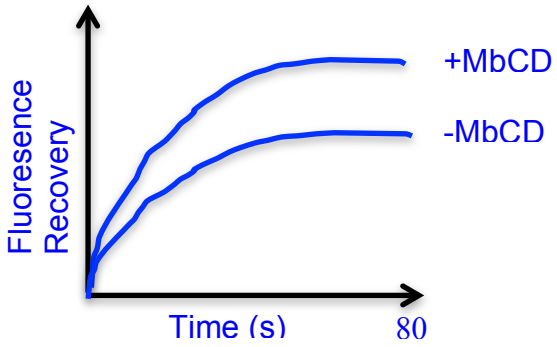
Data adapted from Goodwin et.al. 2005. *Biophysical Journal*. Vol. 89:2 1398-1410

A. Describe what the data in the images above shows, indicate which condition would be considered the control, and explain why. (2 marks)

The data shows that in the control (- MbCD) the Ras-GFP fluorescence recovers slower compared to cells treated with + MbCD, which recovers faster (or moves into the bleached region). (1 mark)

The control is the –MbCD condition (0.5 mark) because treatment with MbCD is the variable, so the untreated condition provides the base of comparison to the treated cells (experimental condition). (0.5 marks)

B. Based on the data, draw what you would expect the fluorescence recovery curves for each condition to look like? Draw and label both curves on the axis below, as well as the x and y axis. (2 marks)



0.5 marks for +MbCD curve
0.5 marks for –MbCD curve
0.5 marks - x axis label
0.5 marks - y axis label

C. Explain what you can conclude about how the mobility of Ras within the plasma membrane may be regulated. (1 mark)

The mobility of Ras is influenced by the presence of cholesterol in the membrane. **Cholesterol restricts the mobility of Ras** (1mark). Other possibility - Ras may be located in cholesterol rich lipid rafts, which limits the mobility of membrane-associated proteins.

(Q4, Part D on next page)

Q4 continued:

D. Predict how the mobility of Ras might differ in an artificial lipid bilayer (made entirely of phospholipids) compared to a cellular plasma membrane? Explain. (2 marks)

The mobility of Ras would be faster in an artificial phospholipid membrane (1 mark) because its lateral diffusion is not restricted by: (name one- 1 mark)
-lipid composition, presence of cholesterol, lipid rafts
-cell cortex, actin cytoskeleton
-cell-cell adhesion
-cell-matrix adhesion

Question 5 (5 marks)

The late Ruth Benerito was a chemist who developed wrinkle-free cotton in the textile industry. Cotton is a polymer of cellulose made up of several long chains of glucose, linked together by H-bonds. Prior to the development of wrinkle-free cotton, washing and tumble-drying regular cotton was enough to wrinkle them so much that they weren't wearable. Clothes need to be ironed before they could be worn. With wrinkle-free cotton, clothes came out of the dryer relatively wrinkle-free and ironing was no longer needed.

A. The formation of 3-dimensional structure in polysaccharides can be considered analogous to the tertiary structure of proteins. Describe the different types of interactions that influence the tertiary structure in proteins, and identify which of these types is the most important in the case of cellulose. (2 marks)

- 1 mark: interactions that influence tertiary structure – ionic, H-bonding, London dispersion, S-S bonds (must have 3 for full mark)
- H-bonding is the most important in the case of cellulose (1 mark)

B. The application of heat in a tumble dryer alters the structure of the cellulose polymers in cotton and causes wrinkles. Explain why heat might change the structure of the cellulose polymer. (1 mark)

H-bonds are broken by heat, which could result in a realignment of the polymers in cellulose.

C. How might you alter the structure of the cellulose polymer to make it wrinkle-free? Provide one example and explain your logic. (2 marks)

Cross-link glucose polymers with covalent bonds, replace the ineffectual hydrogen bonds with stronger ones, more H-bonds. The new bonds stabilize the polymer chains in crisp, unwrinkled form, even in the presence of heat.

Anything that was reasonable, and took into account that cellulose is a polysaccharide (and not a protein, lipid or nucleic acid).

1 mark for example, 1 mark for reasoning.

Question 6 (10 marks)
DNA is an extremely long molecule that must be packed into a tiny nucleus in such a way that the cell can gain access to the genes it needs efficiently. Histones are one of the most important proteins in maintaining the organization of the DNA within the nucleus. Describe the importance of the properties of a histone's amino acids on their non-covalent interactions with each other, and with the other molecules in the histone's environment, that give rise the histone structure and function.

Thesis statement:
Thesis statement should be specific, talk about how protein structure will be influenced by environment as well as function of histones.

Example thesis statement?

This is a good exercise to have them try in groups, and then to have a wider discussion of the strengths and weaknesses of a few of them.

Argument 1 and evidence:
Marking scheme (similar to tutorial):

Organization = 3pts
1pt: thesis statement (0.5 presence, 0.5 quality)
1pt: Presence/ quality of Intro
1pt: Presence/ quality of conclusion (Be generous)
2pts: Overall quality & clarity of essay.

Content = 7pts
The most important task in this outline is to make the connection between the 1) the structure of the histone proteins themselves, 2) the function of histone proteins in DNA packing (and how that function results in specific biochemical properties.

There are a couple ways to go about this.

1. Students could discuss 3 of the 4 levels of protein structure AND how it relates to nucleosome structure.
2. Students could discuss aspects of histone structure (i.e. DNA interactions that are non-permanent, the ability to form the histone octamer) and speak to the specific structural requirements of each one.

Major points for each of the levels of protein structure and how they relate to histone function:

a. Primary:
   - Amino acid sequence will be rich in basic amino acids, as DNA is acidic
   - It will also be rich in hydrophobic amino acids to aid with 3D structure and protein-protein interactions.

b. Secondary and Tertiary:
   - Hydrophobic amino acids on the inside, and in areas where the different histones interact with each other.
   - Basic amino acids around the outside, where the DNA must interact.

c. Quarternary:
   - Nucleosome is an octamer, so 8 different proteins (2x H2A, H2B, H3, H4) must come together in a specific orientation, which will be determined by properties of the surfaces of the histones.
   - Hydrophobic residues for 4° structure ensure that the histones come together correctly, and do not confuse the surfaces meant to interact with other histones with the surfaces that must interact with the negatively charged DNA.

Argument 2 and evidence:

Argument 3 on Next Page

Argument 3 and evidence:

THIS SPACE WILL NOT BE MARKED – USE IT FOR ROUGH NOTES