National Exams December 2018

04-Bio-A3, Cellular and Molecular Biology and Biochemistry

3 hours duration

NOTES:

1. If doubt exists as to the interpretation of any question, the candidate is urged to submit a clear statement of any assumptions made with the answer paper.

2. This is a CLOSED BOOK EXAM.
   An approved Casio or Sharp calculator is permitted.

3. FIVE (5) questions constitute a complete exam paper.
   The first five questions as they appear in the answer book will be marked.

4. Each question is of equal value.

5. This exam contains a question with true and false statements. NOTE: THE MARKING SCHEME FOR THE TRUE AND FALSE QUESTION IS +1 FOR A CORRECT ANSWER, 0 FOR A BLANK STATEMENT, -1 FOR AN INCORRECT ANSWER.

6. Most questions require an answer in essay format. ***Clarity and organization of the answer are important.***

Q	Mark
1	/20
2	/20
3	/20
4	/20
5	/20
Total	/100

Question 1: Protein, gene regulation, model organisms and recombinant DNA technology (20 marks total)

a) **(4 marks)** In your own words, explain what an allosteric protein is.

b) **(5 marks)** In the context of gene regulation, explain how an allosteric protein can behave as a transcriptional repressor.

c) **(5 marks)** Cyclic adenosine monophosphate (cAMP) plays a role in the control of the *lac* operon in *Escherichia coli* (*E. coli*). Explain how cAMP is used to control the transcription of the *lac* operon.

d) **(3 marks)** β-galactosidase, a gene from the *lac* operon, is often used as a reporter protein in biotechnology. Why?

e) **(3 marks)** Explain how the DNA regulatory sequences for the *lac* operon can be used as a biotechnology?

Question 2: Enzyme kinetics and substrate inhibition. (20 marks total)

The rate of a simple enzyme reaction follows Michaelis-Menten kinetics. If the maximum reaction rate (V_{max}) for the enzyme is 85 umoles/sec and the Michaelis-Menten constant (K_M) is 0.8 mM, estimate the reaction rate at a substrate (reactant) concentration of:

a) **(2 marks)** 0.008 mM
b) **(2 marks)** 0.8 mM
c) **(2 marks)** 80 mM

d) **(4 marks)** Justify the above answers by plotting a graph of reaction rate vs substrate concentration and labeling key features (including V_{max} and K_M and the 3 different substrate concentrations)

e) **(10 marks)** Develop an equation for the rate of product formation when there is substrate inhibition. Use an approach that is similar to that used to develop the Michaelis-Menten equation. In the reaction schemes below, E refers to free enzyme, S to substrate, P to product, ES to an enzyme substrate complex, and ESS to a complex involving more than one substrate molecule. Assume that a rapid equilibrium is established between E, S and ES that can be described by the equilibrium constant K_M. Also assume that an equilibrium is established between ES, S and ESS that can be described by the equilibrium constant K_{IS}.

$$E+S \leftrightarrow ES \rightarrow E+P$$
$$ES+S \leftrightarrow ESS$$

Question 3: Antibiotics and antibodies (20 marks total)

Many people confuse antibiotics and antibodies, two very important classes of molecules for the healthcare and pharmaceutical industry. In fact, one such person has come to you for help in identifying whether a solution contains an antibiotic or an antibody.

(20 marks) Devise a set of experiments that would allow you to identify whether the solution contained an antibody or an antibiotic. 10 marks will be awarded for the sets of experiments used to identify if an antibody is present. 10 marks will be awarded for the sets of experiments used to identify if an antibiotic is present.

Question 4: Gene regulation and cell differentiation (20 marks total)

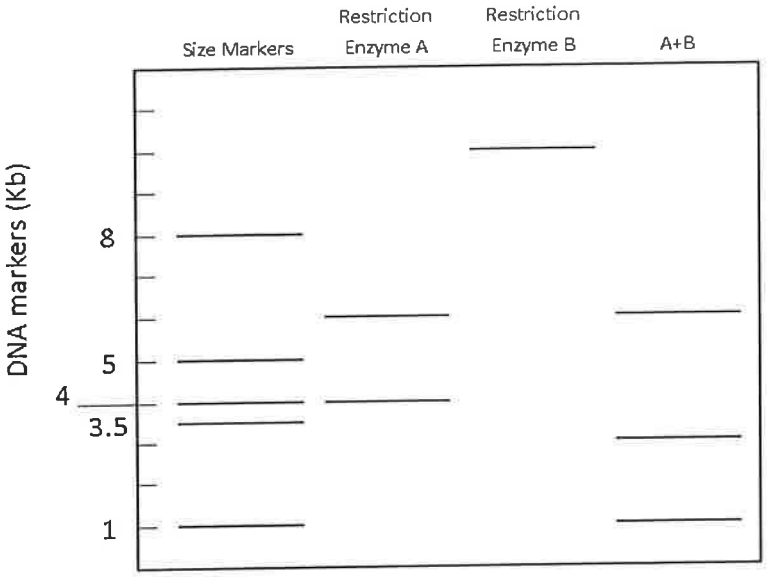
a) **(10 marks)** Explain how the regulation of genes can be passed down from a parent eukaryotic cell to a daughter eukaryotic cell. What are the epigenetic modifications that allow this to happen? Explain how this is related to cellular differentiation.

b) **(5 marks)** What is meant by a positive feedback loop and how could this applied to cellular differentiation?

c) **(5 marks)** What are iPS cells?

Question 5: Restriction enzymes and DNA sequencing (20 marks total)

Your colleague has digested double stranded DNA with two different restriction enzymes (A and B) and the resulting products were analyzed by separating the fragments using gel electrophoresis. DNA fragments of known sizes were electrophoresed on the same gel at the same time as size markers. Questions 5 a, b, and c all refer to the figure below.



a) **(5 marks)** Was the DNA that was digested linear or circular? Explain.

b) **(6 marks)** Estimate the size of each fragment obtained from the 3 different digestions (A, B, A+B).

c) **(5 marks)** From this information, draw a map of the original DNA with the placement (relative position) of the restriction enzyme cleavage sites. Include any size information possible.

d) **(4 marks)** Dideoxynucleoside triphosphates can be used to help identify DNA sequences. How?

Question 6: Cellular and molecular biology and biochemistry (20 marks total)

NOTE: THE MARKING SCHEME FOR TRUE AND FALSE QUESTIONS IS +1 FOR CORRECT ANSWER, 0 FOR BLANK STATEMENT, -1 FOR INCORRECT ANSWER.

1. Hydrogen bonds are weak and can be broken by thermal energy, yet they contribute significantly to the specificity of interactions between macromolecules. _____

2. The active site of an enzyme usually occupies only a small fraction of the enzyme surface. _____

3. Non-covalent bonds are too weak to influence the 3D structure of macromolecules. _____

4. Affinity chromatography separates molecules according to their intrinsic charge. _____

5. Proteins can carry overall negative, positive or no charge depending on the pH of the solution they are in. _____

6. Ultracentrifugation can allow greater separation of small molecules compared to centrifugation at lower gravitational forces. _____

7. If 14% of the nucleotides in the DNA of a bacterial cell are adenine, 36% of the nucleotides in the DNA of that same bacterial cell must be guanine. _____

8. A G-C base pair is stabilized by at most two hydrogen bonds. _____

9. During interphase, DNA is contained in the nucleus of a cell and each chromosome can be easily identified without staining. _____

10. Two closely related species should have the same number of chromosomes. _____

11. Chromosomes, chromatin and chromatids all refer to DNA in association with proteins. _____

12. Chromatids are always present in the cell. _____

13. DNA was determined to be the source of hereditary information through an experiment whereby live virus was able to take up DNA from lysed cells. _____

14. Chemical modification of a histone e.g. methylation of the histone, can regulate the packing of DNA in eukaryotic chromosomes. _____

15. Ion exchange chromatography is used to separate molecules based on charge. _____

16. Sodium dodecyl sulfate (SDS) Polyacrylamide-Gel Electrophoresis (SDS-PAGE) is a technique used to separate proteins based on charge. _____

17. Only the sulfur in cysteine residues can form disulfide bridges (bonds) in proteins. _____

18. In gel-filtration chromatography, proteins are passed over porous material; large molecules are recovered quicker because they are not able to enter the porous material as easily as smaller molecules. _____

19. A protein can have multiple functional domains. _____

20. Antibodies and antibiotics are similar molecules that function in similar ways. _____