University of British Columbia
Mid-term Exam – July 21, 2008
Biochemistry 300 - Summer session

Time: 1.5 hours
Total Marks:100

Candidate's Name: _____
(Please print **family name first.**)

Student Number: _____

Candidate's Signature: _____

This examination consists of 3 parts A, B, and C. Part B has 15 questions and part C has 8 questions. The exam has a total of 11 pages. Please check to ensure that this paper is complete.

Answer all questions on this examination paper in the space provided.

Read and observe the following rules:

1. Each candidate should be prepared to produce, upon request, his/her library/AMS card.
2. No candidate shall be permitted to enter the examination after the expiration of ½ hour, or to leave during the first ½ hour of the examination.
3. Candidates are not permitted to ask questions of the invigilators, (except in cases of supposed errors or ambiguities in examination questions). CAUTION - Candidates guilty of any of the following, or similar, dishonest practices shall be immediately dismissed from the examination and shall be liable to disciplinary action:
   a) Making use of any books, electronic storage devices, papers or memoranda, or cassette recorders other than those authorized by the examiners;
   b) Speaking or communicating with other candidates;
   c) Purposely exposing written papers to the view of other candidates.
   d) The plea of accident or forgetfulness shall not be received.
4. Smoking is not permitted during examinations.
5. Cellular telephones must be fully turned **off**.

Mark Obtained: _____ /100

Equations & Constants:

K = °C + 273
F: 96,480 J/V mol
R: 8.315 J/mol K

$\Delta G' = \Delta H - T\Delta S$

$\Delta G^{o'} = -nF\Delta E_o'$

$\Delta G' = \Delta G'^{o} + RT \ln \frac{[C][D]}{[A][B]}$

$V_o = V_{max}([S]/([S] + K_m))$

$pH = pK_a + \log([A^-]/[HA])$

Where A & B are reactants and C & D are products

$1/V_o = 1/V_{max} + (K_m/V_{max})(1/[S])$

Part A. Matching terms. Chose from the following list below and assign a number. Not all terms will be used. (20 marks)

1. Synthetase
2. $FMNH_2$
3. Entropy
4. Enthalpy
5. Epimers
6. Tertiary structure
7. 3-phosphoglycerate
8. Glyceraldhyde-3-phosphate
9. Activation energy
10. Histidine
11. Valine
12. Dihydroxyacetone phosphate
13. Anomers
14. Leucine
15. Water
16. Gibb's free energy
17. Serine
18. $FADH_2$
19. FAD
20. Holoenzyme
21. Isoleucine
22. Quaternary Structure
23. FMN
24. Apoenzyme
25. Enantiomers
26. Secondary structure
27. Mutase
28. Heme group
29. Synthase
30. Transition state

A. _____ $= G_X^{\ddagger} - G_S$.

B. _____ The spatial arrangement of amino acid residues that are relatively close to one another in the linear sequence.

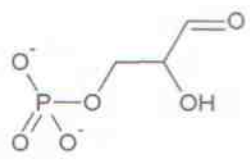
C._____ The heat content of the system.

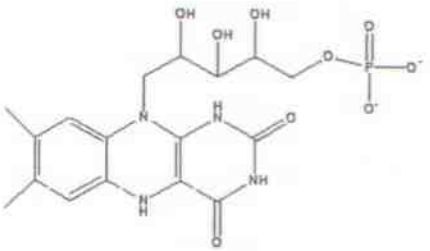
D. _____ Pairs of molecules, each with more than one asymmetric center, that differ in configuration at only one such center.

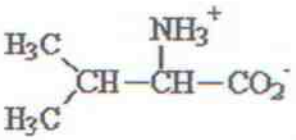
E. _____ Joins two molecules together without ATP (NTP).

F. _____ In the mechanism of chymotrypsin catalysis, it's the second nucleophile.

G. _____ An enzyme with its necessary cofactor(s).

H. _____ 

I. _____ 

J. _____ 

Part B. Multiple Choice. Circle the best answer. (30 marks)

1. What is the $[A^-]/[HA]$ ratio when a weak acid is in a solution 1 pH unit above its pK_a?
   a. 1:1.
   b. 1:10.
   c. 10:1.
   d. 1:2.
   e. None of the above.

2. Which of the following sugars **CANNOT** mutarotate?
   a. Glucose-6-phosphate.
   b. Glucose-1-phosphate.
   c. Fructose-1,6-bisphosphate.
   d. Fructose-2,6-bisphosphate.
   e. b and d.
   f. All of the above.

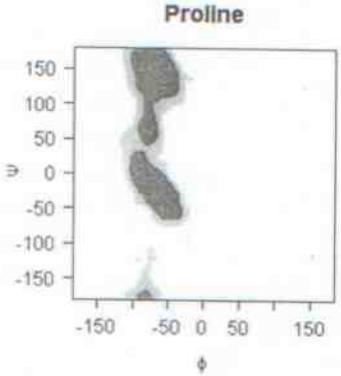
3. The tertiary structure of YFP (Your Favorite Protein) is thought to contain a β-strand on its surface. One side of the strand faces the surface of YFP and the other side faces the core. Which of the following sequences of the β-strand is most likely?
   a. F I M V W G A.
   b. S L E W K M T.
   c. T F V E N V M.
   d. T D S Q E K C.

4. Regarding pathway regulation by ADP, which of the following is true?
   a. It stimulates glycolysis by directly activating hexokinase.
   b. It stimulates glycolysis by directly activating phosphofructokinase.
   c. It stimulates the Krebs cycle by directly activating isocitrate dehydrogenase.
   d. All of the above.
   e. b and c.

5. When the substrate concentration is much greater than K_M, the rate of catalysis is almost equal to:
   a. k_{cat}.
   b. K_M.
   c. V_{max}.
   d. $(V_{max}/K_M)[S]$.
   e. None of the above.

6. Which of the following statements about phosphoglycerate mutase is **NOT** correct?
   a. As part of the mechanism, the enzyme is transiently phosphorylated in the active site.
   b. It can covert 2-phosphoglycerate to 3-phosphoglycerate.
   c. It requires stoichiometric amounts of 2,3-bisphosphoglycerate.
   d. It catalyzes an intramolecular shift of a phosphate group.

7. In one of your initial attempts to determine the tertiary structure of YFP, you predict a proline residue to have dihedral angles of phi = -75° and psi = 125°. Based on the Ramachandran plot for proline below, the predicted orientation of the proline residue is:
   a. Very likely.
   b. Possible, but not optimal.
   c. Not likely.
   d. Unable to be determined.



8. In solution, glucose:
   a. Exists primarily in the furanose form.
   b. Is an aldose.
   c. Is almost exclusively α anomer.
   d. Rapidly converts between D and L forms at room temperature.

9. The steps (in order) to covert succinate to oxaloacetate are:
   a. Oxidation, hydration, oxidation.
   b. Oxidation, decarboxylation, oxidation.
   c. Decarboxylation, oxidation, hydration.
   d. Oxidation, decarboxylation, hydration.

10. Which of the following is **NOT** true concerning glycolysis in anaerobic muscle?
   a. There is a net production of NADH.
   b. There is a net production of ATP.
   c. Lactate dehydrogenase is active.
   d. None of the above (i.e., all of the above are true!).

11. Which of the following statements is true of enzyme catalysts?
    a. Their catalytic activity is independent of pH.
    b. They can increase the reaction rate for a given reaction by a 1000 fold or more.
    c. They can increase the equilibrium constant for a given reaction by a thousand fold or more.
    d. They are generally equally active on D and L isomers of a given substrate.
    e. To be effective, they must be present at the same concentration as their substrate.

12. What would be the expected result of a Lys residue being substituted with a Ser residue in the 2,3-bisphosphoglycerate (BPG) binding site in hemoglobin?
    a. BPG would bind tighter because of the loss of a positive charge.
    b. BPG would bind tighter because of a gain of a positive charge.
    c. BPG would bind less tightly because of the loss of a positive charge.
    d. BPG would bind less tightly because of the gain of a positive charge.

13. In yeast, citrate synthase is a homodimer with two active sites. Through a series of experiments, you have determined that 1 nM of enzyme has a V_{max} of 36 μM/min using citrate as a substrate. Therefore, the turnover number (k_{cat}) is:
    a. 18,000 per minute.
    b. 36,000 per minute.
    c. 72,000 per minute.
    d. None of the above.

14. The pK_a of the carboxyl R group on the amino acid glutamate is 4.1. If the pH is 2, then:
    a. [R-COOH] > [R-COO^-].
    b. [R-COOH] < [R-COO^-].
    c. [R-COOH] = [R-COO^-].
    d. None of the above, the carboxyl group cannot be ionized.

15. The following statements about the regulation of pyruvate dehydrogenase complex are true **EXCEPT**:
    a. NADH activates the kinase that phosphorylates pyruvate dehydrogenase.
    b. Pyruvate inhibits the kinase that phosphorylates pyruvate dehydrogenase.
    c. NADH directly inhibits dihydrolipoyl dehydrogenase.
    d. Acetyl CoA directly inhibits dihydrolipoyl transacetylase.
    e. None of the above (i.e. all are true!).

Part C: Short Answer Questions:

1. Given the following reaction:

1,3-bisphosphoglycerate (1,3BPG) + ADP ⇔ 3-phosphoglycerate (3PG) + ATP

1a. What is the name of the enzyme that catalyzes this reaction? Where in the cell is it found? (2 marks)

1b. Using the table provided below, calculate the $\Delta G^{o'}$ of the above forward reaction. Is this a spontaneous reaction at standard state? Why? (4 marks)

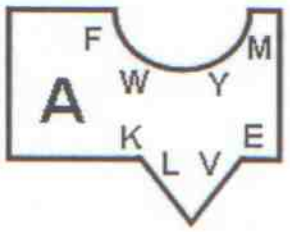
1,3 bisphosphoglycerate + H_2O → 3-phosphoglycerate + P_i	$\Delta G^{o'}$: -49.4 kJ/mole
phosphoenolpyruvate + H_2O → Pyruvate + P_i	$\Delta G^{o'}$: -61.9 kJ/mole
ATP + H_2O → ADP + P_i	$\Delta G^{o'}$: -30.5 kJ/mole
ATP + H_2O → AMP + PP_i (Pyrophosphate)	$\Delta G^{o'}$: -45.6 kJ/mole
3-phosphoglycerate + H_2O → glycerate + P_i	$\Delta G^{o'}$: -10.3 kJ/mole

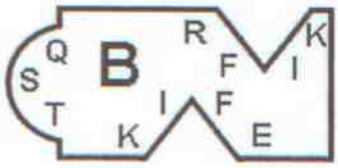
1c. In a fasting individual, the following concentrations were observed in liver cells:

6.0 mM ATP	**0.1 mM 1,3 bisphosphoglycerate**
0.4 mM ADP	**15.0 mM 3-phosphoglycerate**

Using the values above, calculate the ΔG of the forward reaction. Why is this value different than the one calculated in 1b? What is happening with this reaction in liver cells? (5 marks)

2. Proteins can bind to other proteins via a series of weak interactions (in the same way that a substrate can bind to an active site). For the proteins A and B, draw the most likely A:B dimer and comment on the types of interactions that are formed. (4 marks)





3a. An α-helix has a defined structure. Explain how this structure is stabilized/maintained. Be very specific! (4 marks)

3b. Proline residues will cause α-helices to either bend or break. Besides the obvious steric hindrance issues, why would proline residues disrupt α-helices? (3 marks)

4. Papain is a **cysteine** protease that is found in papaya trees. Papain contains three key conserved residues that are essential for catalysis. These are Cys-25, His-159, and Asn-158. Based on your knowledge of the mechanism of chymotrypsin proteolysis, propose a role for each residue. (3 marks)

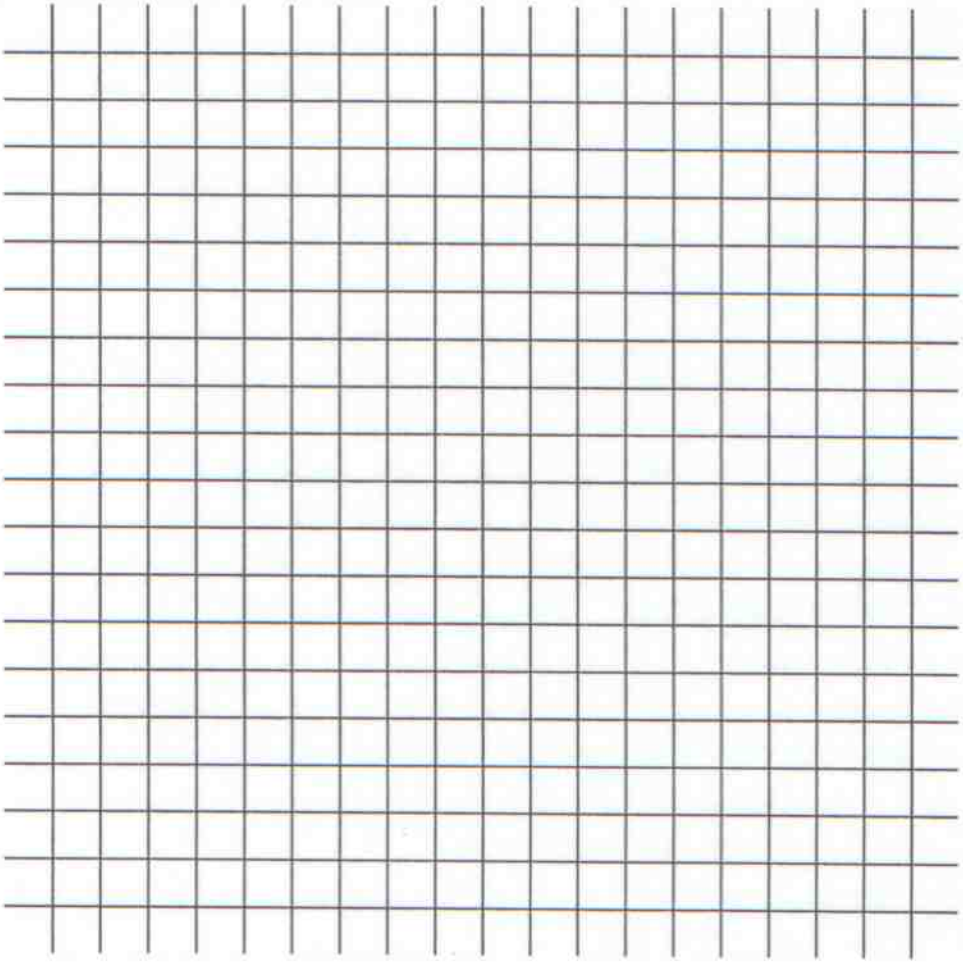
5. 4-hydroxy-2-nonenal (HNE) rapidly and irreversibly reduces Lipoamide. Discuss the effect that HNE has on the α-ketoglutarate dehydrogenase complex. What would be the status of the **bound** co-factors when the complex is treated with HNE? (5 marks)

6. Draw out the **overall** reaction catalyzed by isocitrate dehydrogenase. Make sure to include all names and structures. (4 marks)

7. You are studying an inhibitor of picornavirus RNA polymerase (a cause of the common cold). Through work in your lab, you have confirmed that this enzyme obeys simple Michaelis-Menten kinetics. Furthermore, a post-doctoral fellow has identified a new inhibitor against the RNA polymerase and provided you with this data:

[S] mM	Polymerase V_o (μM/min)	Polymerase + Inhibitor V_o (μM/min)
0.100	1.25×10^{-1}	3.13×10^{-2}
0.125	1.43×10^{-1}	3.57×10^{-2}
0.167	1.67×10^{-1}	4.17×10^{-2}
0.250	2.00×10^{-1}	5.00×10^{-2}

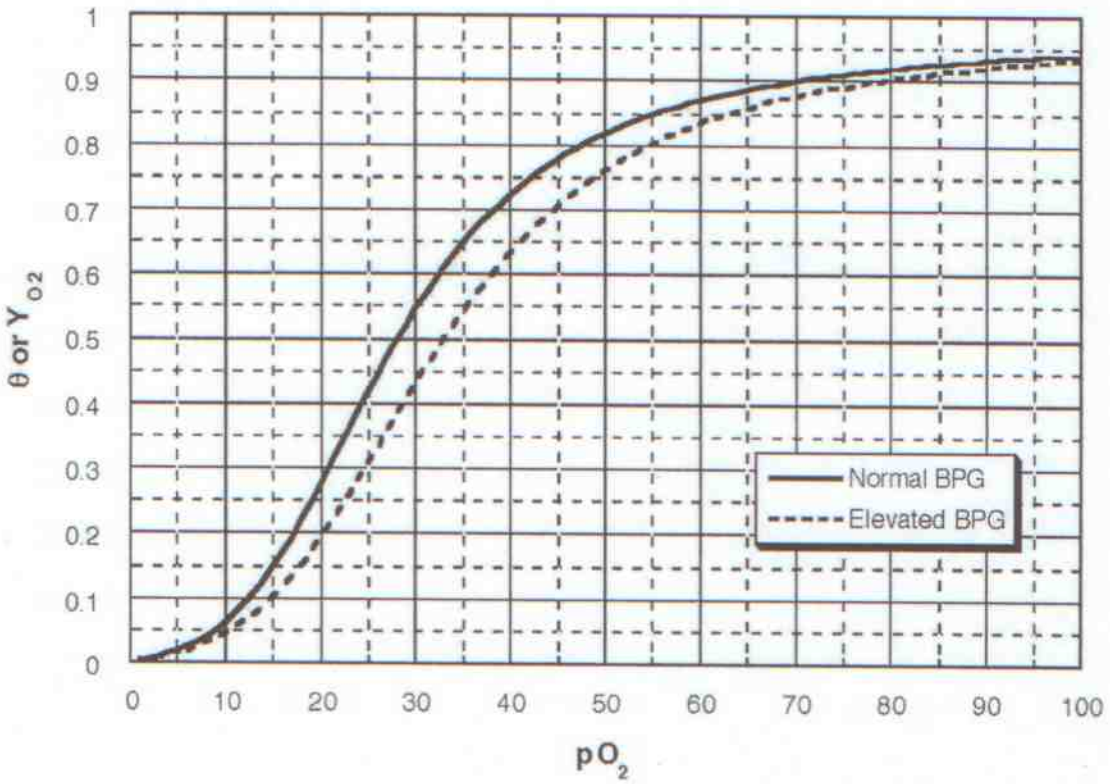
7a. Determine K_m and V_{max} for the Polymerase and the Polymerase + inhibitor. (4 marks)



7b. What type of inhibition is this? How do you know? (3 marks)

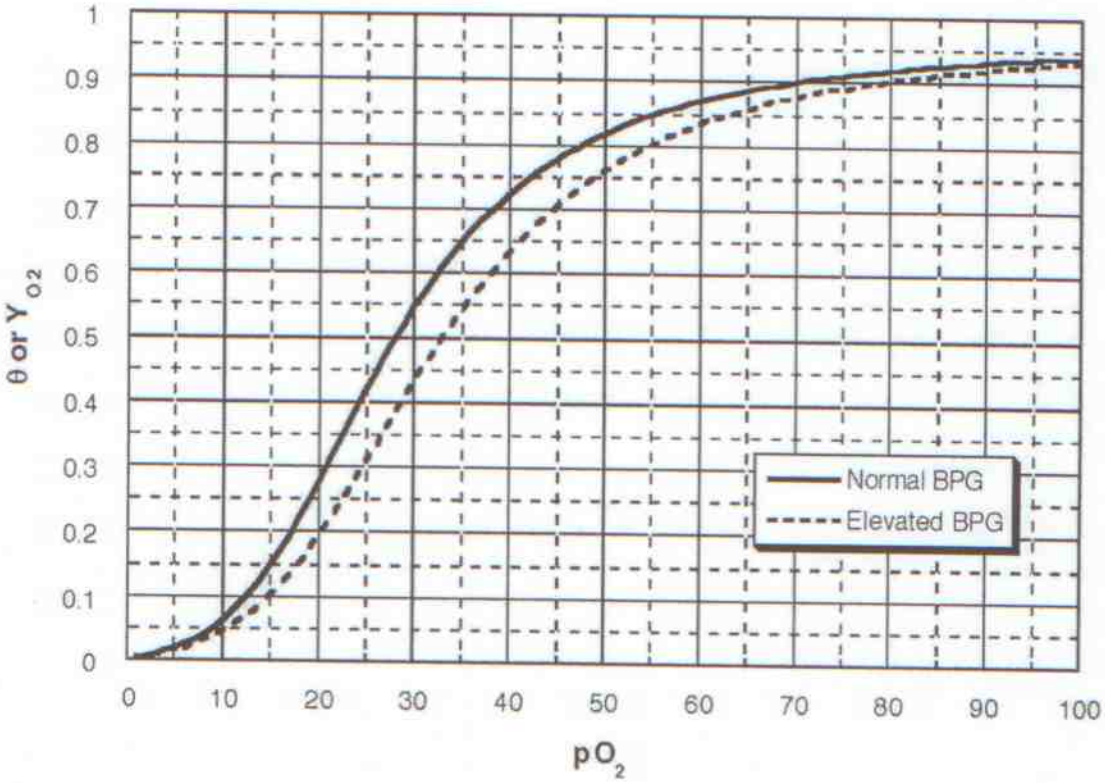
8. You are a geologist studying Mt. Rainier (elevation: 14,400 feet). As part of your work, you spend two weeks on the summit of the volcano. At sea level, pO_2 in the air (and thus the lungs) is approximately 90 Torr, however, at the summit, pO_2 drops to 55 Torr. The pO_2 in tissues is 30 Torr.

8a. In order to acclimatize to the high altitude, your body increases the concentration of 2,3-bisphosphoglycerate (BPG) in red blood cells. The graph (below) shows the O_2 binding curves for hemoglobin with normal (5mM) and elevated (8mM) concentrations of BPG. By drawing directly on the graph, (1) show the effect altitude has on O_2 delivery to tissues with normal concentrations of BPG, and (2) show the effect of increased BPG concentration at high altitude has on O_2 delivery in the tissues. Does an elevated level of BPG help O_2 delivery at high altitudes? (7 marks)



(There is a copy of this graph on the next page in case you mess up)

8b. Oh Smurf! Mt. Rainier is erupting!!!! You're running down the mountain as fast as you can! This results in increased concentrations of CO_2 and H^+ in your muscle tissue. Assuming you still have an elevated BPG concentration, draw the resulting O_2 binding curve for hemoglobin in your muscle on the graph below. (2 marks)



(This is the same graph as on the previous page)