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**BICH 440, EXAM 1, Monday, September 24, 2007, 100 points total**

You MUST sign the following academic integrity statement:

On my honor, I have neither given nor received unauthorized aid on this academic work.

Signed: \_\_\_\_\_

1. Write your name on the cover page ONLY.
2. Write your student UID# on ALL pages. If you do not remember your UID#, make up a random 8 digit number. DO NOT use your social security number.
3. Write concise answers to demonstrate effectively your mastery of the subject. In order to obtain maximum credit, you need to show your work.

gas constant  $R$  8.315 J/mol-K

1.(15 pts) Draw the chemical structure of the peptide KMQR showing the correct ionization at pH6. Draw all peptide bonds to demonstrate atoms in the trans configuration. This peptide is modified such that the amino terminus is acetylated. On your structure show the bond that is cleaved if this peptide is treated with cyanogen bromide.

2.(6 pts) A highly-purified protein sample is electrophoresed on a SDS-polyacrylamide gel both before and after treatment with a high concentration of beta-mercaptoethanol. Before treatment, the sample exhibited three bands with approximate molecular weights of 80,000, 65,000 and 40,000. After treatment with beta-mercaptoethanol, four bands of 80,000, 40,000, 35,000 and 30,000 were detected by electrophoresis. Explain (A); Why does the highly-purified protein sample contain multiple bands on the gel? (B) Why are there four bands after mercaptoethanol treatment vs. three bands before treatment?

3.(5 pts) Your friend, an excellent biochemist, gives you an enzyme sample that she says is absolutely pure. The amount of enzyme is 2 micrograms, and the specific activity is 50,000 Units/mg. Because you are compulsive, you decide to subject this enzyme preparation to one more purification step, which turns out to be a big mistake. You recover 1.5 micrograms of protein, but only one-quarter of the starting overall enzyme activity. What is the specific activity of your final enzyme preparation?

4.(12 pts) Consider the peptide: ITRYHARD

(A) Predict the net charge of this peptide at pH3 and pH8.

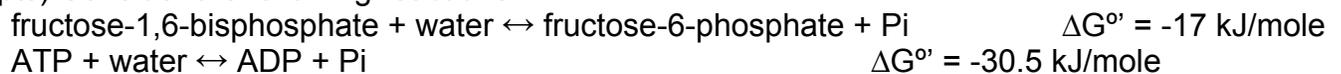
(B) Estimate the isoelectric point for this peptide.

As necessary, use the following pKa values: C $\alpha$  carboxyl: 2.0; sidechain carboxyl: 4.0;

imidazole: 6.0; C $\alpha$  amino: 9.0; sidechain amino: 9.5; phenolic hydroxyl: 10.0; guanidino: 12.5

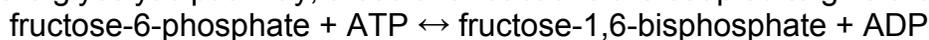
5.(10 pts) You are planning to separate the amino acids arginine, leucine, aspartic acid and glutamine by ion-exchange chromatography. Using an **anion**-exchange polystyrene resin at pH 11.0, predict the order of elution of these amino acids. The primary criterion for separation is charge, and a secondary consideration is hydrophobic character. HINT: Note that the polystyrene resin is hydrophobic. While thinking of your answer, use the pKa's specified in problem 4. REMINDER: Show your work to get maximum credit.

6.(15 pts) Consider the following reactions:



(A) What is the equilibrium constant for the top reaction (37°C)?

(B) In the glycolytic pathway, these two reactions are coupled to give the following reaction:



Calculate the free energy change for this coupled reaction at 37°C when the concentrations of each reactant and product are: [ATP] = 4mM; [ADP] = 1mM; [fructose-6-phosphate] = 0.3 mM; [fructose-1,6-bisphosphate] = 0.2 mM

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7.(15 pts) Starting with 100mL of a 0.5M solution of a buffer called HEPES, pH 7.0, what pH would result after the addition of 10mL of 2M NaOH? The pKa for HEPES is 7.5.

8.(6 pts) Draw the polypeptide backbone, leaving out specific structures of R-groups, for two chains involved in an antiparallel beta sheet. Include three complete residues within each chain, and delineate all interchain hydrogen bonds using dotted lines.

9.(16 pts) Short Answer

- A) Amino acid that has highest molar extinction coefficient for absorption at 280nm
  
- B) Name one amino acid that contains two asymmetric carbon atoms
  
- C) What is the physical basis for separation of proteins by isoelectric focusing?
  
- D) What is the weakest noncovalent interaction?
  
- E) Name one reagent that will react and modify cysteine residues containing free thiols.
  
- F) Approximately how many amino acids in a polypeptide chain of molecular weight 66,000?
  
- G) Approximately how many amino acids exist in an alpha helix that is 2nm long?
  
- H) What famous chemist proposed the structure of the  $\alpha$ -helix found in proteins?