Name _____

I.D. _____

Score _____

Exam 1, BICH 440, Section 500, Monday, September 24, 2001

Write your name on each page. Write concise answers to demonstrate effectively your mastery of the subject. Show your work in order to receive maximum credit where applicable.

gas constant R 8.315 J/mol-K

1) (10 pts) Purification of an enzyme. A biochemist discovers and purifies a new enzyme, generating the purification table below:

procedure	**Total protein (mg)**	**Activity (units)**
1. crude extract	20,000	4,000,000
2. precipitation (salt)	5000	3,000,000
3. precipitation (pH)	4000	1,000,000
4. ion-exchange chromatography	200	800,000
5. affinity chromatography	50	750,000
6. size-exclusion chromatography	45	675,000

A) Complete the purification table, calculating the yields and specific activities for each step.

B) Which of the purification procedures used for this enzyme is most effective (i.e., gives the greatest increase in purity)? Why?
C) Which of the purification procedures is least effective? Why?
D) Is there any indication in this table that the enzyme is now pure? What else could be done to estimate the purity of the enzyme preparation?

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2) (15 pts) Draw the structure of the tripeptide glutamine-methionine-histidine at pH 9. You do not need to depict the stereochemistry of this molecule. Circle the potential hydrogen bond donors in the sidechains of this peptide.

3) (6 pts) The hydrophobic effect can be explained because of an increase in entropy. Why?

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4) (8 pts) Identify the correct amino acid(s):

A) Two amino acids that contain two chiral centers

B) Amino acid with the highest molar extinction coefficient at 280 nm (i.e., it absorbs the most uv light at this wavelength).

C) This amino acid sidechain reacts with an aldehyde to form a Schiff base.

D) This amino acid can form a disulfide bond.

5) (15 pts) Your buffer problem. How much 1M HCl is required to change the pH of 1 liter of 0.1M glycine buffer from pH10 to pH9? The pKs of glycine are 2.3 and 9.6.

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6) (9 pts) Consider the glycolytic reaction,

dihydroxyacetone phosphate (DHAP) $\leftrightarrow$ glyceraldehyde-3-phosphate (G3P)

The equilibrium constant for this reaction is 0.0524 at 37C. The actual free energy change for this reaction in erythrocytes is $\Delta G = +2.4$ kJ/mole. What is the ratio of [DHAP]/[G3P] in erythrocytes at 37C?

7) (12 pts) Consider the peptide: ala-glu-tyr-his-ile. (A) Predict the net charge of this peptide at pH5, pH8, pH12. (B) Estimate the isoelectric point for this peptide. Use the following pKa values: Cα carboxyl: 2.0; sidechain carboxyl: 4.0; sidechain imidazole: 6.0; Cα amino: 9.5; phenolic –OH: 10.0.

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8) (25 pts) Short answer questions.

a) (2 pts) What is the pH of 0.0001M HCl?

b) (2 pts) Name two reagents that react with the thiol group of cysteine.

c) (4 pts) Draw the structure of the sidechain of ε-N-methyl lysine.

d) (3 pts) After what amino acid residues does chymotrypsin cleave a protein?

e) (2 pts) What two electrophoretic techniques are combined to generate two-dimensional protein gels?

f) (4 pts) Briefly explain two reasons why proline is rarely found in alpha helices of proteins.

g) (2 pts) What quantities can be determined from a van't Hoff plot?

h) (2 pts) What system buffers the pH of blood plasma?

i) (4 pts) Draw the stereochemically-correct structure of L-alanine.