

Biochemistry I - First Exam Face Page

This exam has 8 pages, including this one.

Total Points – 100: 2 points/minute

The following equations and constants may be useful:

T=300K and pH=7.0 unless otherwise stated.

R=8.3 J/mol-K RT=2.5 kJ/mol @ 300K

Log2=0.3 ln10=2.3

$$\Delta G^0 = -RT \ln K_{eq}$$

$$\Delta G = \Delta H - T\Delta S$$

$$S = R \ln W$$

$$\ln(a^n) = n \ln a$$

For the reaction: N \rightleftharpoons U:

$$K_{eq} = [U]/[N]$$

$$f_u = K_{eq}/(1+K_{eq})$$

$$f_n = 1/(1+K_{eq})$$

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

$$[HA] = [A_T]/(1+R)$$

$$[A^-] = [A_T]R/(1+R)$$

Beer's law: A=ε[X]l

Amino Acid Names:

Alanine: Ala

Arginine: Arg

Asparagine: Asn

Aspartic Acid: Asp

Cystine: Cys

Glycine: Gly

Histidine: His

Isoleucine: Ile

Lysine: Lys

Leucine: Leu

Methionine: Met

Phenylalanine: Phe

Proline: Pro

Serine: Ser

Threonine: Thr

Tryptophan: Trp

Tyrosine: Tyr

Valine: Val

Glutamine: Gln

Glutamic Acid: Glu

A : _____

B1 : _____

B2 : _____

B3 : _____

B4 : _____

B5 : _____

B6 : _____

C1 : _____

C2 : _____

Tot : _____

Part A: (24 points total: 3 points each, Circle the best answer)

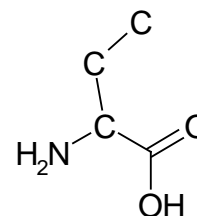
1. The peptide bond in proteins
 - a) is in a tetrahedral configuration.
 - b) is planar, and fixed in a trans conformation.
 - c) is planar, and usually found in a trans conformation.
 - d) can always be cleaved by cyanogen bromide.
2. A monoprotic weak acid can act as an *effective* buffer
 - a) at pH values within one pH unit of its pKa.
 - b) at pH values within two pH units of its pKa
 - c) at pH values within three pH units of its pKa.
 - d) at any pH value.
3. If the ϕ and ψ angles of each peptide unit in a protein are *both* known, the following will also be determined:
 - a) complete secondary structure.
 - b) complete tertiary structure.
 - c) complete quaternary structure.
 - d) Both the first and the second choices are correct.
4. During the unfolding reaction of an α -helix, breakage of each hydrogen bond requires about 2kJ/mol. This implies:
 - a) Hydrogen bonds between groups on the mainchain and water have no intrinsic energy.
 - b) Hydrogen bonds are much stronger in proteins than in water.
 - c) Hydrogen bonds are not reformed with water.
 - d) Hydrogen bonds are slightly stronger in proteins than in water.
5. The Standard Gibb's free energy, ΔG° , is
 - a) the amount of energy required to convert 1 mole of reactant to its equilibrium concentration.
 - b) the residual energy present in the product(s) at equilibrium.
 - c) the energy required to convert one mole of reactants to one mole of products
 - d) the difference in the residual energy of reactants and products at equilibrium.
6. Which of the following is most correct:
 - a) Charged amino acids are never buried in the interior of a protein.
 - b) Charged amino acids are seldom buried in the interior of a protein.
 - c) All hydrophobic amino acids are buried when a protein folds.
 - d) Tyrosine is only found in the interior of proteins.
7. Which of the following is the most unfavorable for protein folding?
 - a) Conformational Entropy.
 - b) Hydrophobic Interactions.
 - c) Van der Waals Interactions.
 - d) Hydrogen Bonds
8. Antigens bind to which portions of an antibody?
 - a) variable regions.
 - b) constant regions.
 - c) hypervariable regions
 - d) only heavy chains.

Part B: Short Answer

B1 (7 pts): An α -amino acid is shown to the right (this may or may not be one of the common 20 amino acids found in proteins.).

i) Clearly Label the α -carbon with the Greek letter " α ".(1 pt)

ii) Convert this amino acid into one of the 20 common amino acid that has a polar and uncharged sidechain by using the smallest number of alterations to the structure. Sketch the amino acid in the space below.(3 pts)

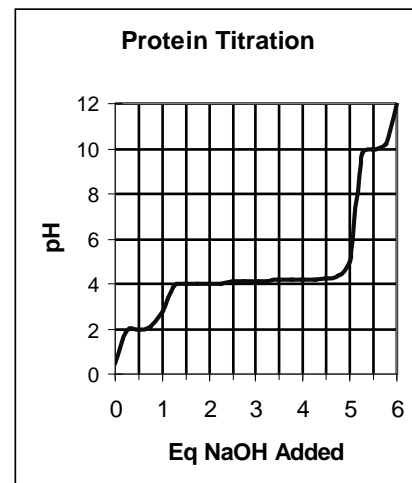


iv) Show how one water molecule could form a hydrogen bond with the sidechain of the amino acid that you just sketched. Label the donor or acceptor atoms on the sidechain.(3 pts).

B2:(8 pts) The pH titration curve of a 50 residue protein is shown in the diagram to the right. This protein contains only one type of an ionizable amino acid.

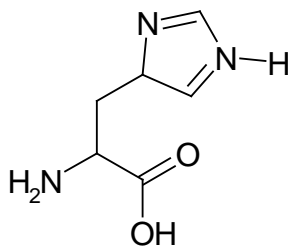
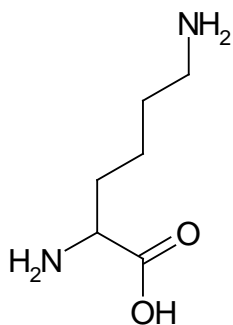
i) Based on this titration curve, circle the ionizable amino acid from the following list: Asp His Lys? Briefly justify your choice.(4 pts)

ii) How many ionizable residues of the type you identified in part *i* are present in this protein? Why? (2 pts)

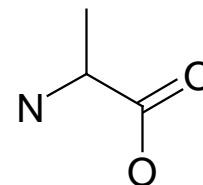


iii) *Estimate to within two pH units* the isoelectric point (pI) of this protein. Justify your answer. If you are short on time, tell me how you would approach the problem.(2 pts)

B3.(10 pts) The following are the structures of two amino acids that are ionizable under biological conditions:



i) Pick one of the two amino acids and sketch its predominate ionic form at pH=8.0 using the diagram shown to the right *or* the diagram given above. You can assume that the pK_a values of the amino and carboxy termini are 9.0 and 2.0 respectively. Be sure to write down the pK_a that you have assumed for the side chain, draw hydrogen atoms on ionizable groups where necessary and indicate charges when necessary.(5 pts)



ii) Draw the dipeptide in an extended chain conformation that would result from forming a peptide bond between these two amino acids; you need not draw the sidechains (just label them R1 and R2) but you need to draw all mainchain atoms. (4 pts)

iii) Write the name of your dipeptide (Sample answer: Ala-Phe)(1 pt)

B4: Do **only one** of the following three parts to this question (10 points):

B4a: Discuss the major changes in *entropy* that occur when a protein folds into a typical compact globular protein. Be sure to discuss which bonds along the mainchain of the polypeptide backbone are involved with the entropy change.

OR

B4b: Briefly discuss how the environment of an ionizable group can affect its pK_a value. Provide an example using any ionizable group found in proteins.

OR

B4c: Compare and/or contrast the “forces” associated with protein folding and the binding of antigens (proteins, carbohydrates, or haptens) to immunoglobulins.

B5:(7 pts)

Briefly discuss the hierarchy of protein structure (e.g. primary, secondary, tertiary, quaternary) using immunoglobulins, or any other example, in your discussion.

B6: (12 pts) The handout for this exam shows the structure of a small peptide. The oxygen atoms are colored red, the nitrogen atoms colored blue, the hydrogen atoms are colored white. Carbon atoms are colored light-gray in the sidechains and dark grey for those in the mainchain. Note that not all hydrogens are shown. **Please write your name on the page** and then answer the following questions:

i: What is the secondary structure of this peptide?(3 pts)

ii: Could this peptide be found on the surface of a larger protein, or is it more likely to be buried within the non-polar core? Briefly justify your answer.(2 pts)

iii. Sketch a water molecule forming a hydrogen bond with the amide proton or the carbonyl oxygen of the third residue in the sequence of this peptide.(2 pt)

iii: Assuming that this peptide were part of a larger protein, list two super-secondary structures that this segment could participate in.(2 pts)

iv) The following is a list of structural features in this peptide. Write the appropriate letter from the diagram adjacent to the structural feature. The fourth entry has been done as an example.(1/2 point each)

____:A hydrophobic Amino Acid

____:A peptide bond

____:A ψ angle

____:A ϕ angle

____:The Carboxy-terminus

____:The amino-terminus

A :An amino acid that strongly absorbs UV light

Part C: Detailed Calculations

C1 (10 pts): Do only one of the following four parts, please indicate the question you are attempting.

i) Given the following monoprotic acids with the following pK_a values:

Acetic acid: 5.0 Imidazole: 6.0 Tris: 8.0

Briefly describe how you would make 1L of a 0.1M solution that will be a good buffer at pH 4.0. State which of the above acids you would use and why. You only need to calculate the number of moles of the compound(s) required to make the buffer.

OR

ii) A protein contains two Trp ($\epsilon_{280} = 5000 \text{ M}^{-1}\text{cm}^{-1}$) residues and one Tyrosine residue ($\epsilon_{280} = 1000 \text{ M}^{-1}\text{cm}^{-1}$). A solution of this protein has an absorbance of 2.0 for a 1cm path length. What is the concentration of the protein in solution?

OR

iii) An enzyme uses a cysteine residue within its active site to accomplish its chemistry. This cysteine residue has a pK_a of 7.0 and must be fully deprotonated for full activity. What is the % activity of the enzyme at pH= 6.0.

OR

iv) A 8 residue peptide was treated with trypsin, yielding two peptides of length 3 and 5. The sequence of the five residue peptide was: Ala-Gly-Met-Thr-Lys.

The peptide, when treated with chymotrypsin, produced two peptides, length 2 and 6. The tube containing the 6-residue long sequence was dropped on the floor, thus it was only possible to obtain the sequence of the 2 residue peptide: Leu-Leu.

Determine as much of the sequence of this peptide as possible (Hint, you may not have enough information to completely determine the sequence).

One last question on the next page:

C2:(12 points)

You work for Bampbell's soup company, in their process control division. Botulism toxin is a deadly poison that is occasionally found in canned goods. Fortunately, the toxin is a protein and can be inactivated by heat. Consequently, you would like to add a high temperature step to the canning process to insure that the canned soup is toxin free. The chefs tell you that temperatures above 87 C (360 K) will destroy the delicate flavor of their soup and they implore you to keep the temperature below 87 C.

The ΔH° and ΔS° for denaturation of botulism toxin are +360 kJ/mol and +1000 J/mol-K, respectively (Watch the units, enthalpy is in kJ/mol, entropy is in J/mol-K).

i) Given that you need to inactivate 99% of the toxin, can you meet the wishes of the chefs and keep the temperature during detoxification below 360 K? Please show your work and justify your answer ($R=8.3$ J/mol-K).(9 pts)

ii) Regardless of the outcome to part i, can you suggest a way to reduce the temperature of the detoxification process and still denature the toxin?(3 pts)