

Name: \_\_\_\_\_

## Biochemistry I-First Exam

This exam has a total of 90 points and is divided into three sections. You must do ALL of the questions in sections A and B. You should only do two of the three questions in section C. Clearly indicate which questions in section C you want to have graded. Otherwise the first two (C1 and C2) will be graded.

There are a total of 13 pages in this exam. Before beginning please write your name on every page before you begin and use the space provided to answer the question.

Grade: \_\_\_\_\_

Part A: \_\_\_\_\_

B1: \_\_\_\_\_

B2: \_\_\_\_\_

B3: \_\_\_\_\_

Ca: \_\_\_\_\_

Cb: \_\_\_\_\_

The following equations and constants may be useful:

$T=300\text{K}$  and  $\text{pH}=7.0$  unless otherwise stated.

$R=8.3\text{ J/mol}\cdot\text{K}$      $RT=2.5\text{ kJ/mol @ }300\text{K}$

$\text{Log}2=0.3$      $\ln10=2.3$

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

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

$S = R\ln W$

$\text{pH} = \text{pK}_A + \log([A^-]/[HA])$

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

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

$A = \epsilon[X]l$

Name: \_\_\_\_\_

1. The partial negative charge at one end of a water molecule is attracted to the partial positive charge of another water molecule. This attraction is called:
  - a) a covalent bond.
  - b) a hydration shell.
  - c) a hydrogen bond.
  - d) a hydrophobic bond.
2. Which of the following alcohols would be most insoluble in water?
  - a) methanol ( $\text{CH}_3\text{OH}$ )
  - b) ethanol ( $\text{CH}_3\text{CH}_2\text{OH}$ )
  - c) butanol ( $\text{CH}_3\text{CH}_2\text{CH}_2\text{CH}_2\text{OH}$ )
  - d) octanol ( $\text{CH}_3[\text{CH}_2]_6\text{CH}_2\text{OH}$ )
3. The amino acid Threonine, can act as an acid-base buffers
  - a) at both alkaline and acid pHs
  - b) at pH values around neutrality.
  - c) at pH values halfway between its  $\text{pK}_a$ 's.
  - d) at pH values around any of its  $\text{pK}_a$ 's.
4. Which pair of amino acids absorbs the most UV light at 280 nm?
  - a) Threonine & Histidine
  - b) Tryptophan & Tyrosine
  - c) Cystine & Aspartate
  - d) Glycine & Tryptophan
5. Which of the following is not a sensible grouping of amino acids based on their polarity properties?
  - a) Ala, Leu, and Val.
  - b) Arg, His, and Lys.
  - c) Phe, Trp, and Tyr.
  - d) Asp, Ile, and Pro.
6. The isoelectric point of an amino acid is defined as
  - a) the pH where the molecule carries no electric charge.
  - b) the pH where the carboxyl group is uncharged.
  - c) the pH where the amino group is uncharged.
  - d) the pH of maximum electrolytic mobility.
7. The peptide bond in proteins is
  - a) planar, but rotates to three preferred dihedral angles.
  - b) polar, but rotates to three preferred dihedral angles.
  - c) nonpolar, and fixed in a trans conformation.
  - d) planar, and usually found in a trans conformation.
8. Hydrogen bonds in  $\alpha$ -helices are
  - a) not present at Phe residues.
  - b) analogous to the steps in a spiral staircase.
  - c) roughly parallel to the helix axis.
  - d) about 5 Å in length.

Name: \_\_\_\_\_

9. The Gibb's free energy,  $\Delta G$ , is negative for
- non-spontaneous processes.
  - spontaneous processes.
  - temperature-independent processes.
  - none of the above.
10. The most important conclusion from Anfinsen's work on denaturation and refolding Ribonuclease (RnaseA) was that:
- the conformation the native conformation of a protein is adopted spontaneously.
  - disulfide bonds (S-S) in proteins can be reduced with b-mercaptoethanol.
  - Urea is not an effective reagent for protein denaturation.
  - 100% enzyme activity corresponds to the native
11. Which of the following is *most* correct:
- Charged amino acids are never buried in the interior of a protein.
  - Charged amino acids are seldom buried in the interior of a protein.
  - All hydrophobic amino acids are buried when a protein folds.
  - Tyrosine is only found in the interior of proteins.
12. Which of the following 'forces' is the most favorable for protein folding?
- Conformational Entropy.
  - Van der Waals Interactions.
  - Hydrogen Bonds
  - None of the above.
13. Unpaired H-bond donors and acceptors are found in the hydrophobic core of a protein
- only at the ends of  $\alpha$ -helices.
  - only at the turns connecting  $\beta$ -strands.
  - only on Pro residues.
  - only very rarely.
14. Structures of anti-PCP antibody shows that the tight binding of PCP to the antibody is *primarily* due to:
- hydrophobic 'forces'.
  - electrostatic forces.
  - van der Waals forces.
  - hydrogen bonding.
15. Monoclonal antibodies produced in the laboratory
- lack light chains.
  - cannot be used for disease diagnosis yet.
  - derive from human cancer patients.
  - can be selected to bind to almost any known molecule.

Name: \_\_\_\_\_

B1. Select **one** of the following amino acids:

Asparagine (Asn), Glutamine (Gln), Tyrosine (Tyr), Tryptophan (Trp).

1. Draw the missing part of the amino acid sidechain.
2. Indicate potential hydrogen bond donors and acceptors for this amino acid.
3. Draw two water molecules in your diagram, one acting as a hydrogen bond donor and one as an acceptor

B2: You have been asked to prepare 1 L of a 0.1 M buffer solution with a pH of 7.0. This buffer will be used to control the pH of a reaction that generates acid. The following buffers are available:

Buffer	pKa
Tris	8.3
Tes	7.5
Mops	7.2
Pipes	6.8

i) Which buffer would you select? Why?

ii) How many moles of the acid (HA) and conjugate base ( $A^-$ ) would you use to make this buffer solution?

iii) After carefully making this buffer, a fellow lab worker comes by and tells you that he thinks the balance that you used to weigh out the acid and base was not working properly. You check the pH of your solution and it is spot on, so you know the ratio of acid to base is correct. Describe how you would experimentally determine the total concentration of the buffer if you are given a solution of 1 M NaOH. A simple figure may be the best way to answer this question.

B3: Of the following amino acids: Ile, Met, Val, Ala

i) Which of the following amino acids has the lowest solubility in water?

ii) Support your answer with a sketch of the amino acid you chose in part i, indicating the part(s) of the amino acid that will not interact favorably with water.

Name: \_\_\_\_\_

B4: The extinction coefficient for Prot G is 10,000 L/M-cm. When protein G unfolds its extinction coefficient changes to 11,000 because the Trp and Tyr residues become exposed to the water in the denatured (unfolded) form of the protein.

i) A solution of native, or folded, Protein G has an absorbance of 1.0. What is the concentration of Protein G in this solution?

ii) If this solution is heated to a temperature well above the  $T_M$  for denaturation, such that all of the protein G is denatured, what absorbance would you measure?

iii) What absorbance would be measured when the temperature is equal to  $T_M$ ?

iv) The following absorbance measurements were obtained at the following temperatures:

Temp.	Abs. Measurement
308K (35 C)	1.00
318K (45 C)	1.00
328K (55 C)	1.01
338K (65 C)	1.03
348K (75 C)	1.08
358K (85 C)	1.10

a) Subtract the absorbance at  $T=298$  from all of the above values and plot this difference on the graph given next to the data table.

b) On the left-hand axis of this plot, draw the correct scale for the fraction unfolded.

c) Indicate the  $T_M$  on this plot.

d) Given a  $\Delta S^0$  value of 600 J/mol-deg, estimate the change in enthalpy due to unfolding. briefly show how you arrived at your answer.

v) A mutant protein unfolds at the same  $T_M$  as the wild-type protein, but the  $\Delta S^0$  of unfolding for this mutant is 2 fold less than that of the wild-type.

a) Is the  $\Delta H$  for unfolding of the mutant lower or higher than that of the wild-type protein? Why?

b) Sketch the transition (melting curve) for the mutant protein on the graph above. Keep in mind your answer to the previous question.