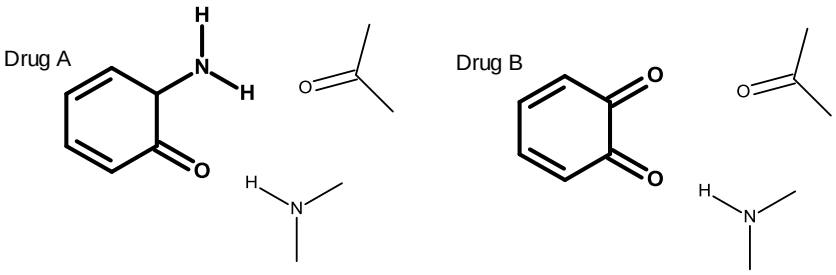
Instructions: This exam consists of 100 pts, 14 questions, 5 pages. On questions with choices all of your attempts will be graded and you will receive the best grade. **Allot 1 min/2 points.**

1. (6 pts) The following shows two drug that are bound to a protein. The structure of the drug is in bold.

i) label **either** a hydrogen bond donor or an acceptor on **drug A**. (4 pts)

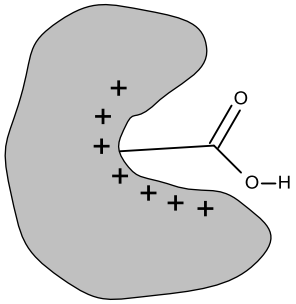
ii) Which of the two drugs would have a lower K_D ($=k_{off}/k_{on}$)? Drug A or drug B? Briefly justify your answer by reference to k_{ON} and k_{OFF} (2 pt).



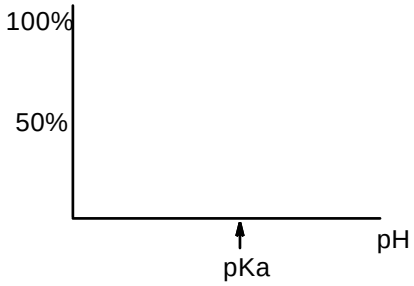
2. (2 pts) Why are most salts soluble in water, even though the ions may interact less favorably with the water molecules than in the crystal.

3. (8 pts) The normal pKa for a carboxylic acid side chain in an amino acid is 4.0. You are measuring the pKa of the same group, but incorporated into a protein. This residue is located in a positively charged pocket (see sketch on the right).

i) Do you expect the pKa to be higher or lower than 4.0 for this residue in the protein? *Briefly justify your answer* (4 pts).



ii) Assume that the **de**protonated species is the active form of this protein, sketch the activity versus pH on the plot to the right (4 pts).



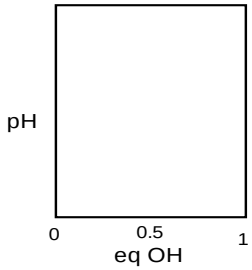
Points on this Page:_____

4. (16 pts) You wish to make a solution that is buffered against pH changes. The desired pH of the solution is 4.0. The volume is 0.5L and the total concentration of the weak acid is 0.1 M The following monoprotic buffers are available: pyruvate (pKa=2.0), acetate (pKa=5), Tris (pKa=8).

$R=10^{pH-pKa}$, $f_{HA} = 1/(1+R)$, $f_{A-} = R/(1+R)$

i) Which of these would you select and why (2 pt)?

ii) Why does the weak acid reduce (buffer) pH changes when a strong acid or base is added to the solution, i.e. explain how a buffer works. A sketch of a titration curve may be useful to illustrate your answer. Feel free to use the diagram on the right (6 pts).



iv) Please do **one** of the following two choices (8 pts).

Choice A: Describe how you would make the buffer – i.e. give moles of HA & H, and the number of moles of NaOH or HCl that would be required, depending on the method you selected to make your buffer.

Choice B: Although the starting pH of your solution was 4.0, it has shifted to 5.0 due to the production of base by the reaction. How many moles of HCl do you add to return the pH to 4.0?

5. (10 pts) Draw the chemical structure of a dipeptide that contains any two amino acids that are **different**. *Do not use alanine or threonine.* Assume that the pH of the solution is 2.0 and indicate the correct protonation state of any ionizable groups. You can assume the relevant pKa values are 2.0 and 9.0 (8 pts).

i) Indicate the sequence of your peptide (1 pt).

ii) *Estimate* the charge on your peptide, at pH=2.0 (1 pts).

Points on this Page:_____

6. (6 pts) Please do **one** of the following choices:

Choice A: Is there free rotation about the peptide bond? Why or why not?

Choice B: The atoms that participate in the peptide bond, O=C-N-H, all lie in a plane, why?

7. (10 pts) The sequence of an 11 residue peptide is determined by Edman sequencing of fragments that are produced after cleavage by CNBr and Chymotrypsin. You should assume that it is possible to only obtain the first five (5) residues of any peptide during the sequencing reaction.

CNBr Fragments:	Ala-Gly-Met	Ala-Ala-Leu	Phe-Arg-Ser-Trp-Met
Chymotrypsin fragments:	Ala-Gly-Met-Phe	Arg-Ser-Trp	Met-Ala-Ala-Leu

Reconstruct the original sequence of the peptide, write your answer here (the first three are done for you).

Ala	Gly	Met								
1	2	3	4	5	6	7	8	9	10	11

Justify your approach to receive partial credit in case your answer is wrong.

8. (3 pts) Circle the interactions that stabilize secondary structure. Put a box around those which stabilize tertiary structure. ~~Strike out~~ any that have no effect on either secondary or tertiary structure.

a) van der Waals

b) Hydrogen bonds

c) Electrostatic interactions

d) Hydrophobic effect

Points on this Page:_____

9. (8 pts)

i) Sketch **one** regular secondary structure and indicated the location of the sidechains and Hydrogen bonds (6 pts).

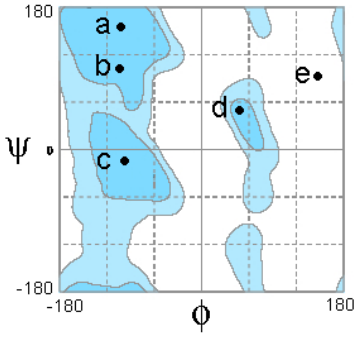
ii) If the secondary structure that you drew faced the core of the protein, what type of side chains would point into the core and how often would you find them in the sequence? (2 pts)

10. (1 pt) How many residues are in one turn of an α-helix?

11. (8 pts) The following is a Ramachandran plot. The values for five residues (a-e) have been plotted. Please answer the following questions.

i) What do the contour lines/shaded areas represent? (6 pts)

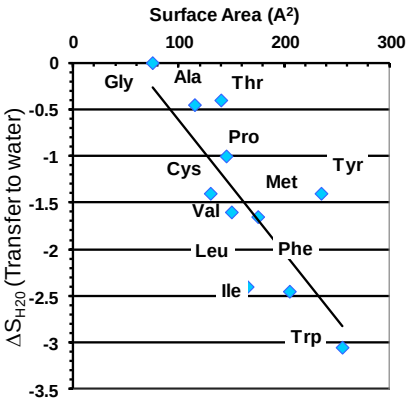
ii) Which of the five points most likely corresponds to a glycine residue? Briefly justify your answer. (2 pts)



12. (11 pts) In the equation: $\Delta S^0 = \Delta S^0_{CHAIN} + \Delta S^0_{SOLVENT}$

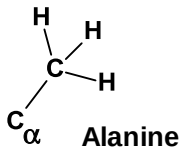
i) What does the term $\Delta S^0_{SOLVENT,}$ represent with respect to protein unfolding? Your answer should include a brief description at the molecular level (8 pts).

ii) *Briefly* describe how you could calculate, or estimate, **either** of the two terms, ΔS^0_{CHAIN} **or** $\Delta S^0_{SOLVENT.}$ The chart on the right may be helpful, as might this equation: S=R ln W (3 pts).

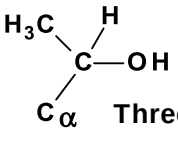


Points on this Page:_____

13. (6 pts) An alanine residue is found in the core of a folded globular protein (wild-type protein). It is replaced by a threonine residue (mutant protein). The amount of unfolded protein for the mutant (threonine) is measured as a function of temperature and plotted on the right. A van't Hoff plot from this data is also on the right. The slope of the line is indicated on the plot.



Alanine



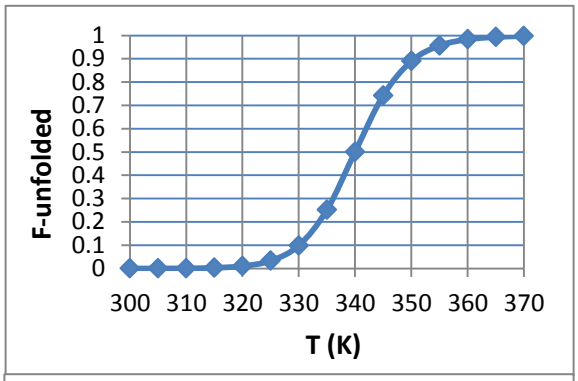
Threonine

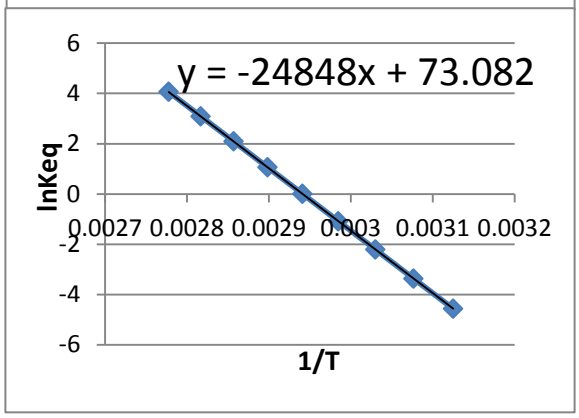
$\Delta G^o = \Delta H^o - T\Delta S^o$

$\ln K_{EQ} = -(\Delta H^0/R)(1/T) + (\Delta S/R)$

R=8.31 J/mol-K

The ΔH^o for unfolding of the wild-type protein is +200 kJ/mol. The entropy for unfolding, ΔS^0, is +600 J/mol-K for ***both*** proteins.





Please do **both** parts *i* and *ii*, you have a choice for part *ii*.

i) Determine the enthalpy for unfolding of the mutant (threonine) containing protein. You can either use the van't Hoff plot, or use T_M = 340K (3 pts).

ii) Please do **<u>one</u>** of the following choices (3 pts):

Choice A: Explain the difference in enthalpy between the wild-type and the mutant protein in terms of interactions that have either stabilized or destabilized the mutant protein.

Choice B: Why is the entropy the same for both proteins? The graph on the previous page may be useful?

14. (5 pts) Please do **<u>one</u>** of the following choices – *use the back of the previous page if you need more space.*

Choice A: Describe, or draw, either an intact antibody, Fab fragment, or F_V fragment. Your answer should describe the separate chains that make up the protein and you should also indicate where the antigen binds and the location of the hypervariable loops.

Choice B: Why do disulfide bonds stabilize the folded structure of antibodies?

Choice C: A protein contains 2 tryptophan (Trp) residues and one tyrosine (Tyr) residue. Write an expression that would allow you to calculate the extinction coefficient for this protein, given that you know ε_{Trp} and ε_{Tyr}.

A=⌈X⌉ε⌊

Points on this Page:_____