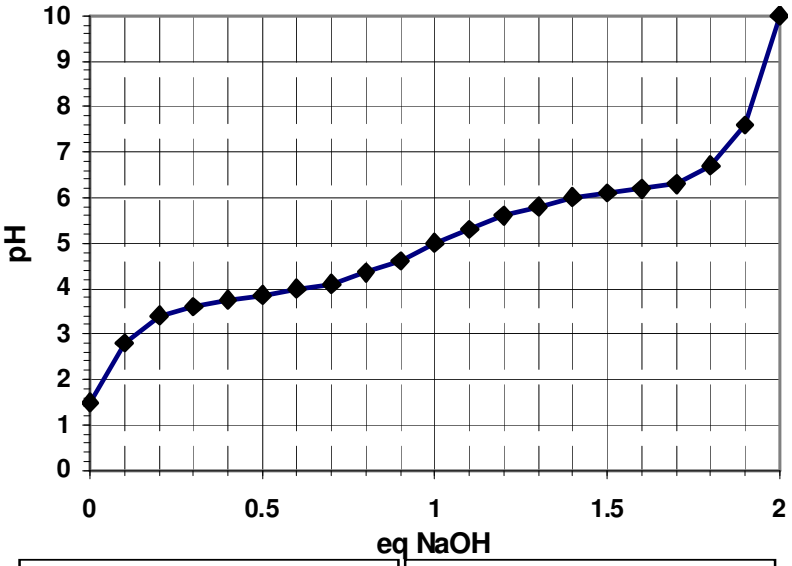
This exam contains **100 pts** in **12 questions** on **5 pages**. Please use the space provided. **Allot 1 min/2pts**.

1. (8 pts) A titration curve for a dipeptide is shown to the right. In this dipeptide the termini have been chemically modified and can no longer ionize; only the sidechain groups can ionize.



i) (1 pt) The pK_a value for the first ionization is 3.8, what is the pK_a for the second ionization?

ii) (2 pts) What ionizable amino acids are contained in this peptide? [Hint, read choice B of the next question.] Write your answers here →

iii) (5 pts) *Briefly* explain why the pH doesn't change very much around pH 4 or pH 6, as base is added.

2. (12 pts) Please do ***one*** of the following two choices. [Hint: The equations to the right are not really required to do this problem.]

$$pH = pK_a + \log\frac{[A^-]}{[HA]}$$

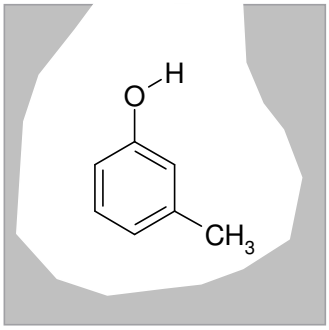
$$f_{HA} = \frac{1}{1+R}$$

$$R = 10^{pH-pKa}$$

Choice A: Describe how you would make 2 liters of a 0.1 M buffer solution at pH 4.0 using the dipeptide discussed in the previous question. Assume that the only source of the weak acid is the fully protonated form, H_2A (i.e. you are starting at the left of the titration curve. Remember that the termini are modified and do not ionize. Show your work.

Choice B: Determine the charge at pH 3.8 for the peptide discussed in the previous question. You may assume that the more acidic residue has a neutral charge when protonated, and the other a positive charge when protonated. Remember that the termini are modified and do not ionize.

3. (8 pts) The diagram to the right shows a compound bound to a protein. The grey area represents the protein.



i) (4 pts) Based on the structure of the bound compound, what forces are likely to be responsible for its binding to the protein? Clearly label the functional groups on the compound (e.g. "a", "b", etc.) and *briefly* discuss how they could interact with the protein in the space below:

ii) (4 pts) If one of the interactions that you identified in *part i* was removed, how would the dissociation constant (K_D) change? Would it increase, decrease, or stay the same? Why?

4. (7 pts) Please do ***one*** of the following two choices.

Choice A: A 10 residue peptide is being sequenced. The partial amino terminal sequence is: Ala-Gly-Met-Phe The peptide is treated with a cleavage reagent and the resultant peptides are separated and their individual sequences are:

Peptide A: Phe-Leu-Lys-Met Peptide B: Asp-Ala-Met Peptide C: Ala-Gly-Met

i) (5 pts) Determine the sequence of the peptide and write the sequence in the space below. Briefly describe how you arrived at your answer.

Ala	-	Gly	-	Met	-	Phe	_____	_____	_____	_____	_____	_____
1		2		3		4	5	6	7	8	9	10

ii) (2 pts) Circle the name of the reagent that was used to cleave the peptide: ***Trypsin*** ***Chymotrypsin*** ***CNBr***

Choice B: A solution of a protein with 5 tyrosine residues and one tryptophan residue has an absorbance of 0.5 at 280 nm.
i) What is the concentration of the protein in solution (4 pts)?
ii) What assumption did you make to solve this problem (3 pts)?

$$A = C\varepsilon l$$
$$\varepsilon_{280}^{Tryptophan} = 5{,}000 M^{-1} cm^{-1}$$
$$\varepsilon_{280}^{Tyrosine} = 1{,}000 M^{-1} cm^{-1}$$

5. (8 pts) Sketch one regular secondary structure in the space to the right. Your drawing should provide:
   i) A representation of the mainchain shape.
   ii) An indication of the location of hydrogen bonds.
   iii) An indication of the location of the sidechains.
   iv) The name of the structure you have drawn.

6. (12 pts)
   i) Draw a dipeptide using any two different amino acids, *except for* phenylalanine and valine (6 pts)
   ii) Provide the sequence for your peptide (e.g. Phe-Val) (2 pts)
   iii) Identify the peptide bond (2 pts)
   iv) Identify all bonds corresponding to the phi (φ) and psi (ψ) torsional angles (2 pts)

7. (8 pts) Please do ***one*** of the following two choices:

Choice A: Describe/draw the most common conformation of the peptide bond and explain why this conformation is low in energy (You may refer to the above drawing, question 7.)

Choice B: Briefly describe a Ramachandran plot and explain why there are three regions of low energy for most amino acids.

8. (8 pts) Please answer ***one*** of the following three choices. Be sure to indicate your choice.

Choice A: Briefly describe the major thermodynamic factor that *destabilizes* the native (folded) state of a protein. Use an equation if appropriate.

Choice B: Explain what thermodynamic factor(s) are responsible for the fact that most proteins have well packed cores.

Choice C: The energy to break a hydrogen bond is approximately 20 kJ/mol, yet hydrogen bonds have a relatively minor role in stabilizing folded proteins, on the order of 1-5 kJ/mol. Why is this so?

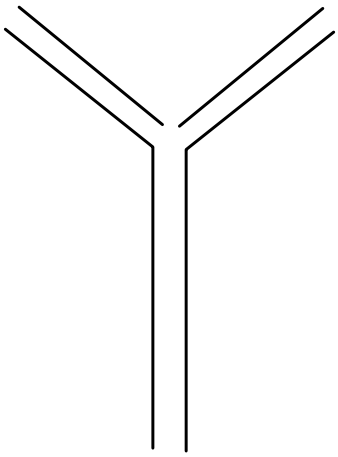
9. (8 pts) Describe the hydrophobic effect in molecular terms and explain why it stabilizes the tertiary structure of proteins but not isolated secondary structures.

10. (3 pts) A simple drawing that represents the structure of an entire antibody molecule is shown on the right. Indicate on this diagram:

i) The location of the variable regions.

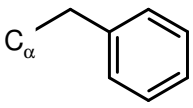
ii) The location of the antigen binding site.

iii) The region of the antibody that would be found in an F_{AB} fragment.

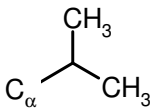


11. (2 pts) Provide one example of the use of antibodies.

12. (16 pts) The thermodynamic stability of four proteins is compared, the wild-type sequence, two proteins with single amino acid replacements, and one protein with both replacements. The locations of these changes are with the core of the protein. The enthalpy and entropy of denaturation are given below and the structures of the sidechains of phenylalanine (Phe) and valine (Val) are shown on the right.



Phe



Val

	Wild-type	Phe57 → Val	Val98 → Phe	Phe57 → Val Val98 → Phe
ΔH^o	+198 kJ/mol	+190 kJ/mol	+170 kJ/mol	+198 kJ/mol
ΔS^o	+600 J/mol-deg	+610 J/mol-deg	+590 j/mol-deg	+600 J/mol-deg

i) (4 pts) Determine how much of the wild-type protein is folded at 330 K. Please show your work.

$$\Delta G = \Delta H - T\Delta S$$
$$\Delta G^o = -RT \ln K_{EQ}$$
$$f_{Native} = \frac{1}{1 + K_{EQ}}$$
$$(N \rightarrow U)$$
$$R = 8.31 j/mol - \text{deg}$$

ii) (4 pts) Sketch, in the space to the right, the denaturation curve for the wild-type protein. Be sure to accurately represent the midpoint of the curve.

iii) (2 pts) *True or False?*

The enthalpy, ΔH^o, is obtained from the slope of the denaturation curve, at T_M. Circle the correct answer above.

iv) (6 pts) Complete ***one*** of the following choices. Use the back of the preceding page if you need more space.

Choice A: Based on the changes in enthalpy (ΔH^o), what can you say about the relative location of residues 57 and 98 within the core of the protein? Are the next to each other, or far apart? Briefly justify your answer.

Choice B: Explain the difference in entropy (ΔS^o) between the wild-type protein and the Phe57→Val mutant, i.e. why is the overall entropy change larger for the mutant protein.

Do not write in this box.