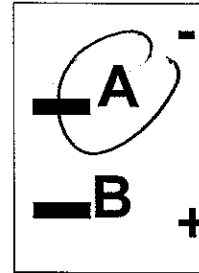


total = 80

10

(2 points) Ion exchange chromatography separates proteins on the basis of Charge while gel filtration separates proteins on the basis of size.

(1 point) The diagram to the right is a schematic of an SDS PAGE gel showing the position of 2 proteins, A and B. Also shown are the negative and positive ends of the gel. Which protein has the larger molecular weight? Circle one in the diagram.



(2 points) You have 2 peptides with the following sequences.

Arg-Glu-Ala-Leu-Ile-Ser-Leu-Asp-Gln-Tyr-Glu Met-Phe-Gln-Asn-Val-Thr-Ile-Leu-Ala-Gln-Arg

You want to separate these 2 using chromatographic methods at neutral pH. Would you use gel filtration or ion exchange chromatography? Why. 1 point for correct answer and 1 point for correct reason.

Both are about the same size so gel filtration is no good. ~~The~~ First peptide has a charge of -2, second +1 so ion exchange is the best

(2 points) You are purifying a protein and find that the initial specific activity is 10 μ moles per second per mg of protein. That is, the enzyme converts 10 μ moles of substrate to product per second per mg of enzyme solution. Suppose 20% of the enzyme solution is the enzyme you want while the remaining 80% is proteins with no enzyme activity. What will be the specific activity of the 100% pure enzyme? No partial credit.

Pure enzyme will have 5 times the specific activity as the initial solution. Answer is 50 μ moles per sec per mg

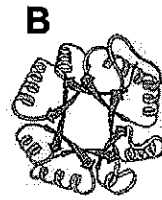
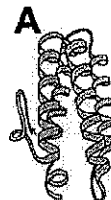
(3 points) 1 point for each correct answer

Protein(s) A has/have all helices.

Protein(s) D has/have all sheets

Protein(s) C & B has/have a mixture of sheets and helices

Write the correct letter A, B, C, or D in the space provided. There may be more than one protein per answer.



15 pts 10 questions

(3 points) A particular peptide has the following amino acid composition (Lys, Phe, Met, Asp, Thr). The following experimental observations have been made.

- Trypsin yields 2 peptides with composition (Lys, Met, Asp) and (Thr, Phe).
- Cyanogen bromide cleavage gives 2 peptides with composition (Met, Asp) and (Lys, Thr, Phe).
- Chymotrypsin gives 2 peptides with composition (Lys, Met, Phe, Asp) and (Thr).

What is the amino acid sequence? Use the 3 letter amino acid code. No partial credit

From (a) we know Met-Asp-Lys or Asp-Met-Lys

From (b) we know Asp-Met so (a)+(b) = Asp-Met-Lys

From (c) we know Phe-Thr so sequence must be Asp-Met-Lys-Phe-Thr

(1 point) The peptide shown below has a net charge of -2 at neutral pH.
+ Asn-Asp-Arg-Glu-Ser-Gln-Val-Leu-Asp -

(1 point) A lipid bilayer surrounding a cell is about 30Å thick. In order for an alpha helix to span this distance, a minimum of 20 amino acids are needed? $1.5 \text{ \AA per residue} \times 20 = 30$

(1 point) The amino acid Pro cannot serve as an H-bond donor in a helix or sheet.

(1 point) Let P=polar amino acid while H=hydrophobic amino acid. Write down the sequence of a hexapeptide that would be consistent with an anti-parallel beta sheet. PHPHPH or HPHHPH

(1 point) Why is there no freedom of rotation around a peptide bond? Can be answered in one sentence. peptide bond has partial double bond character

(2 points) You have discovered a new protease that only cleaves the peptide bond between X and Y (X-Y) where X is an amino acid with a chiral side chain. 3 peptides will be generated by treating the following peptide with your new protease. No partial credit.

Phe-Glu-Thr-Gln-His-Leu-Val-Thr-Ser-Trp-Tyr-Leu-Leu-Cys

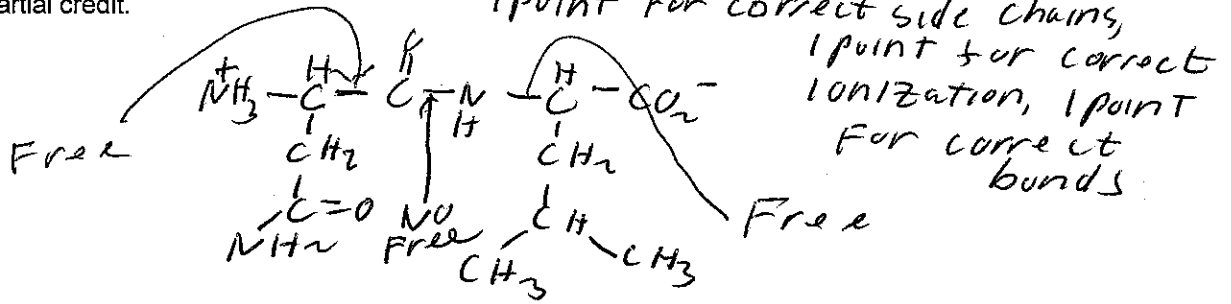
(2 points) In the RNase folding experiment the S-S bonds are broken with a reducing agent and the protein denatured in urea. In order to recover 100% enzyme activity you must first remove urea

followed by remove reducing agent and/or allow S-S bonds to form

(1 point) In an alpha helix the carbonyl O atom of amino acid 1 accepts an H-bond from the peptide NH group of amino acid 5.

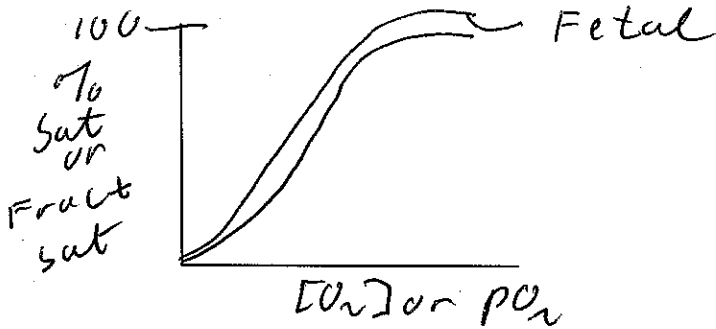
(2 points) A common folding pattern observed in many proteins is called the TIM barrel where a series of parallel sheets are surrounded by helices. A particularly stable form of the TIM barrel is when 8 beta strands are surrounded by 8 helices. Write down two numbers in the spaces provided.

(3 points) Write the structure of the dipeptide Asn-Leu at pH 7. Indicate which bonds along the polypeptide chains there is freedom of rotation and which bonds there is no freedom of rotation. Partial credit.

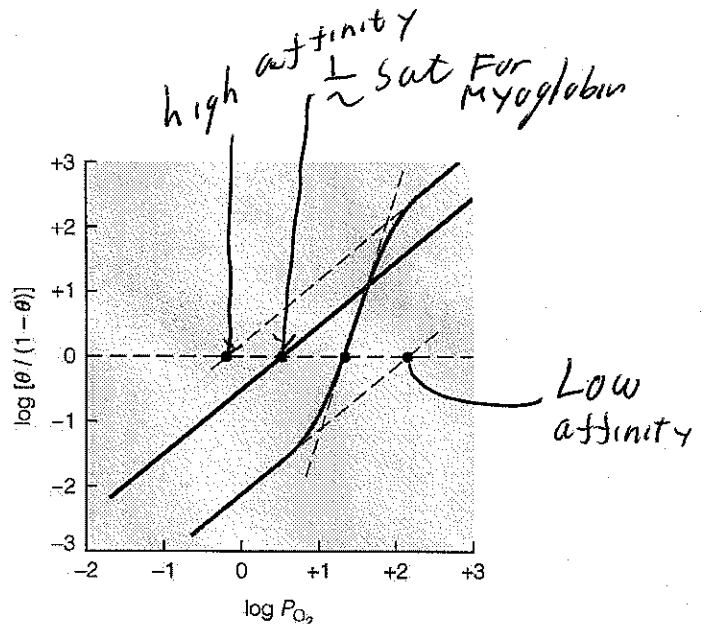


(2 points) In the T to R state transition in hemoglobin, the IRON moves into the porphyrin plane which also causes the amino acid HIS coordinated to the iron to move. Use the 3 letter abbreviation of the amino acid

(3 points) Fetal hemoglobin has a higher affinity for O₂ than adult hemoglobin. Sketch the O₂ binding curves for both types of hemoglobin and indicate which is which. The plot is O₂ concentration vs. fractional saturation. Label the axes. 1 point for correct curves, 1 point for correct labeling, 1 point for identity of curves:



(2 points) The figure is a typical Hill plot for oxygen binding to myoglobin and hemoglobin. A) Indicate the point of half-saturation for myoglobin. B) Indicate the point of half saturation for the low affinity and high affinity forms of hemoglobin.



16

(2 points) There are 2 different models for allosteric enzymes and proteins sometimes called the "sequential" and "concerted" models. In the concerted model there are 2 forms of hemoglobin while for the sequential model there are 5 forms.

(3 points) The binding substrate S to the enzyme E has an equilibrium constant $K_{eq} = 10^4 M^{-1}$. You know that the binding of S to E and the dissociation of the ES complex is fast compared to the conversion of the ES complex to product. V_{max} for this enzyme is $24 \times 10^{-4} M/sec$. What will the velocity of the reaction be at a substrate concentration of $10^{-4} M$? Providing just the correct answer gets full credit. Setting up the correct equations gets 1 point.

$$K_m = K_D = \frac{1}{K_{eq}} = 10^{-4}$$

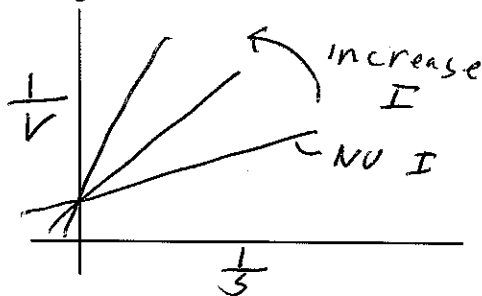
$$V = \frac{V_{max}(S)}{K_m + (S)} = \frac{(24 \times 10^{-4})(10^{-4})}{10^{-4} + 10^{-4}} = \frac{24 \times 10^{-8}}{2 \times 10^{-4}} = 12 \times 10^{-4} M/sec$$

(2 points) The role of the oxyanion hole in serine proteases is to stabilize the negative charge on the carbonyl O atom of the substrate. O to draw C=O

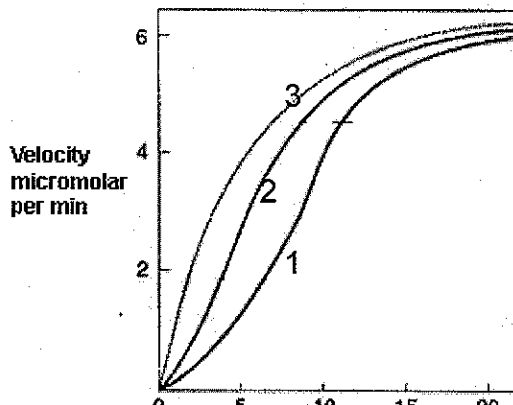
(2 points) In serine proteases the active site amino acid Ser forms a covalent bond with the substrate and the active site amino acid His is an acid-base catalyst.

(1 point) An enzyme lowers the energy of the Transition State/activated complex

(3 points) Label the axis for a Lineweaver-Burke plot. Draw 3 lines: one at 0 inhibitor concentration and 2 other lines at 2 different inhibitor concentrations. Indicate which line corresponds to the higher inhibitor concentration. Assume competitive inhibition. Partial credit given.



3 points



You have a heterodimeric allosteric enzyme consisting of a catalytic domain and a regulatory domain. The regulatory domain binds a positive regulatory molecule A leading to positive cooperativity. The regulatory domain also binds a negative regulatory molecule B leading to negative cooperativity.

Curve 2 has neither A or B in solution

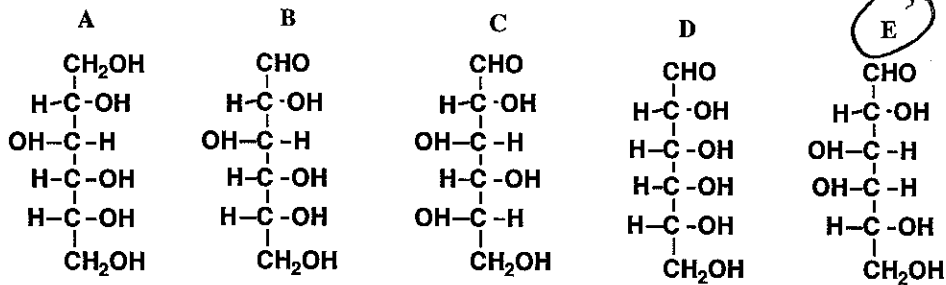
Curve 3 is in the presence of A.

Curve 1 is in the presence of B.

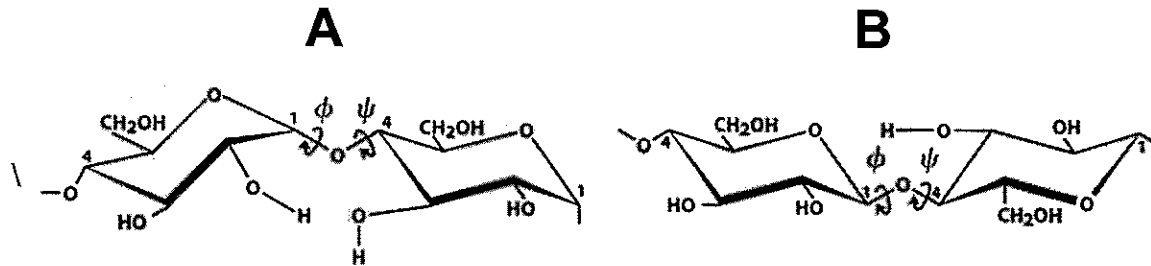
(3 points) The product of the aspartate carbamoyltransferase (ATCase) reaction is CTP (abbreviation OK) which also is a feedback inhibitor of ATCase activity while purines are feedback activator of ATCase activity

(1 point) What type of post-translation modification leads to the activation of chymotrypsinogen to chymotrypsin? One word a short sentence is enough. proteolysis

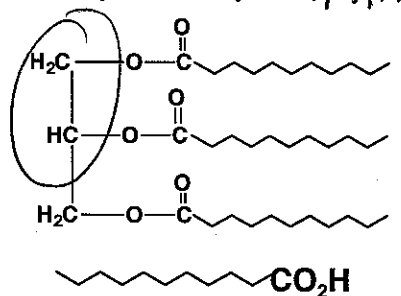
(1 points) Which of these sugars is the 4 epimer of glucose? Circle the correct answer.



(2 points) Amylose and cellulose are both polymers of glucose that use 1,4 linkages. The diagram below shows two types of 1,4 linkages, one for amylose and one for cellulose. The linkage shown in B is the type of linkage found in cellulose. Which of the 2 polymers, cellulose or amylose, forms a more rigid structure? cellulose.



(3 points) The 2 structures shown are different forms of fatty acids. Circle the one that is used to store fatty acids. Why? 1 point for correct choice, 1 point for each correct reason



Answer in this space

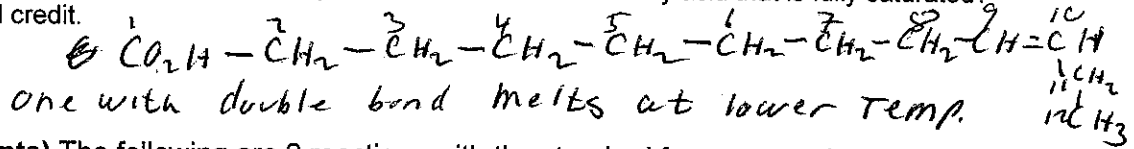
Does NOT Form Micelles and is less soluble.

13

(3 points) The oxidation of hexanol and hexane to give carbon dioxide and water are both very favorable reactions with negative ΔG values. The oxidation of hexane gives the more negative ΔG . Why? 1 point for filling in the blank correctly and 2 points for correct reason.

hexane is more reduced & has more electrons or H atoms. More energy released upon oxidation

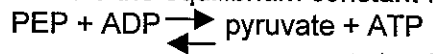
(2 points) Draw the structure of a 12 carbon fatty acid that has a 9,10 double bond. Will this fatty acid melt at a higher or lower temperature than a 12 carbon fatty acid that is fully saturated?
Partial credit.



(2 points) The following are 2 reactions with the standard free energy change

Reaction	ΔG° kcal/mol
$ \text{PEP} + \text{H}_2\text{O} \rightleftharpoons \text{pyruvate} + \text{Pi} $	-14.79
$ \text{ADP} + \text{Pi} \rightleftharpoons \text{ATP} + \text{H}_2\text{O} $	+7.29

What is the equilibrium constant for the following reaction at room temp. (~~30000~~)



we first add to 2 ΔG values to get -7.5 kcal/mol

$$\ln K = -\Delta G / RT \quad \& \quad K = e^{-\Delta G / RT} = e^{7500 / 1.98(300)} = \cancel{3 \times 10^5} \quad 304,450 \text{ or } 3 \times 10^5$$

(2 points) The very favorable hydrolysis of ATP is used to drive unfavorable biochemical reactions while NADH stores the electrons generated during the photosynthetic oxidation of water to oxygen.

(1 point) One positively charged atom and one negatively charged atom are at a distance of 3.0 Å with an electrostatic energy of interaction = -8 kcal/mol. If the atoms are separated by a distance of 6.0 Å the energy of interaction is -4 kcal/mol

(3 points) You have an enzyme with a non-polar substrate binding pocket that is exposed to solvent and a non-polar substrate. When the two form an ES complex the binding $\Delta G < 0$. The release of water is one reason why formation of the ES complex is favorable. This release leads to a increase in which thermodynamic parameter? entropy

(2 points) What is the pH of a solution of 0.12M NH_4Cl and 0.03M NaOH (pK_a of $\text{NH}_4^+/\text{NH}_3$ is 9.25)? No partial
 Problem 14 chap 2 Answer is 8.7 or 8.8

3

(3 points) Sketch the titration curve for the amino acid alanine and indicate on the curve the pKa values and what type of group is being titrated. You must label the X-axis and indicate the 2 pKa values as well as what group is being titrated. No need to give the pKa values. 1 point for correct labeling, 1 point for correct identity of pKa, and 1 point for correct curve.

