CHEM 527 SECOND EXAM FALL 2004

YOUR NAME: _____

NOTES:

1. where appropriate please show work - if in doubt show it anyway.
2. pace yourself - you may want to do the easier questions first.
3. please note the point value of questions - adjust your answers and effort accordingly.
4. some questions may have more data than you need.
5. please be brief - unfocused, rambling answers won't receive as much credit as a few short appropriate phrases.
6. Please write CLEARLY - if I cannot read it - it is wrong.
7. Good luck

Question 1. (29 pts.) Short problems. Show work, but most credit goes to the correct numerical answer. Some questions contain more data than you will need.

a. A pure rat enzyme shows a rate of 2.6 μmol/min for a certain concentration of substrate at 20 °C. The substrate shows a K_m of 3 mM and a molecular weight of 280 g/mol. At saturating substrate concentration the rate is 8.55 μmol/min. What is the substrate concentration?

[S] = _____mM

b. In "a" above the molecular weight of the substrate and enzyme were 280 and 48,000 g/mol respectively and the amount of enzyme used was 40 μg. What is the maximal turnover number (at substrate saturation) at 20 °C?

TN = _____/min

Calculate, or discuss, what turnover number would be expected at 80 °C.

TN = _____

c. Tetrameric human hemoglobin has a total molecular weight of 64,000 g/mol. The atomic weight of iron is 56 g/mol. A human contains 500 g of hemoglobin. Calculate (no partial credit)

how many moles of oxygen can be bound by 500 g hemoglobin _____mol

how many moles of DPG (BPG) can be bound by 500 g hemoglobin _____mol

how many grams of iron are there in 500 g of hemoglobin _____g

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d. No partial credit. If a SINGLE human glycogen molecule contains 120,000 monosaccharide units. About how many:

i.) monosaccharide units in this molecule have their "4" position NOT ATTACHED via a glycoside linkage to another monosaccharide unit

number _____

ii) monosaccharide units have their "1" position NOT ATTACHED to another monosaccharide unit

number _____

iii) monosaccharide units have their "3" position NOT ATTACHED to another monosaccharide unit

number _____

e. An enzyme has a Km of 20 mM for substrate S. What concentration of the substrate would give 90% of the maximal rate?

[S] = _____mM

f. The Km for a substrate is observed to be 38 μM in the presence of 1.6 mM of a competitive inhibitor. In the absence of the inhibitor it is 3 μM. What is the K_i for the inhibitor.

K_i = _____M

g. The reaction: A + B ↔ C + C

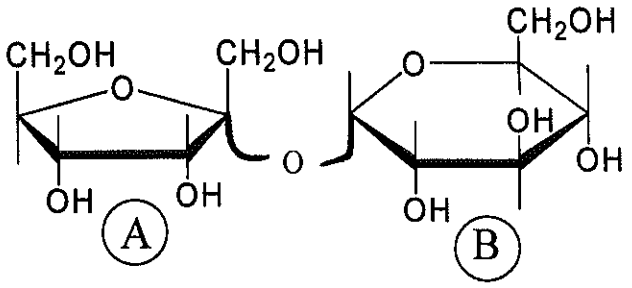
shows a standard free energy change of -4 kcal. Calculate the free energy change for the reaction at 300 °K using these concentrations: A = 10 mM; B = 7 mM; and C = 0.01 mM. The gas constant is 2 cal/degree/mol.

ΔG = _____kcal

The original solution is diluted by 10-times. What is the new free energy change

ΔG = _____kcal

Question 2 (12 pts.) For the disaccharide shown to the right:



a. Name the glycosidic bond:

_____

b. Circle the anomeric carbon in ring B

c. Give the molecular formula (e.g. $C_2H_6O_1$) for the disaccharide

C_____H_____O_____

d. Number carbon atoms 1,3 and 6 in ring A

e. After hydrolysis of the glycoside, ring A is a (circle one)

ketohexose ketopentose aldohexose aldopentose none of these

and ring B is a (circle one)

ketohexose ketopentose aldohexose aldopentose none of these

Question 3 (9 pts) Short answers. Just a few phrases. Please stay within the area allotted.

a. **Normal paper loses most of its strength when wet with water but retains most of its strength when wet with olive oil. Clearly explain the basis for this..**

b. **Human red blood cells contain a single membrane. Carefully describe a method that would allow you to find that 80% of the phosphatidyl-ethanolamine was located on the inner leaflet and only 20% on the outer leaflet of the bilayer.**

c. **An enzyme is reported to have a k_{cat} of 80,000/sec and a Km for its substrate of 0.5 μM. Briefly comment.**

Question 4 (5 pts.) What is the effect of the following on hemoglobin. Circle the most appropriate answer. NC = no change

Increasing pH on the oxygen affinity of hemoglobin	*increase*	*NC*	*decrease*
Increasing oxygen concentration on CO_2 binding to hemoglobin	*increase*	*NC*	*decrease*
Decreasing DPG on oxygen affinity of hemoglobin	*increase*	*NC*	*decrease*
Increasing DPG levels on CO_2 affinity of hemoglobin	*increase*	*NC*	*decrease*
Decreasing pH on the ratio of [T]/[R] state of hemoglobin	*increase*	*NC*	*decrease*

Question 5 (3 pts) What is the most appropriate answer?

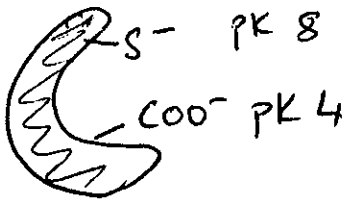
a. the higher the K_m the higher the affinity
b. at [S] = 2 K_m , v = V_{max}
c. at [S] = 2 K_m , doubling the enzyme concentration would exactly double the rate
d. K_m is exactly one half of the maximal velocity
e. all of the above are false.

Question 6 (3 pts) What is the most appropriate answer?

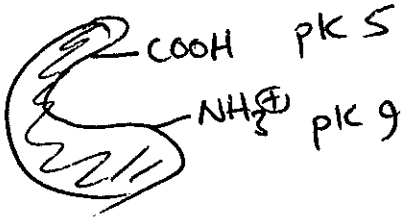
a. bacteria raise their temperature by increasing the proportion of unsaturated fatty acid chains in their membranes
b. Increasing the percentage of unsaturated chains helps keep reindeer legs close to bulk body temperature.
c. unsaturated chains are only found in the inner leaflet of biological bilayers
d. cholesterol is not found in mammalian cell membranes
e. all of the above are false

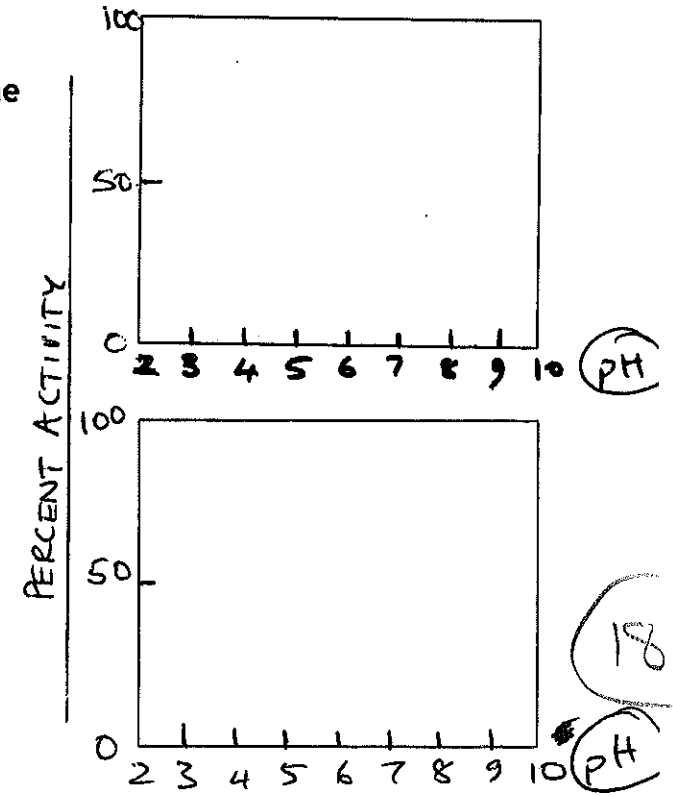
Question 7 (9 pts.) Draw the pH activity curves for the following situations.

a. Only this protonic form is active.

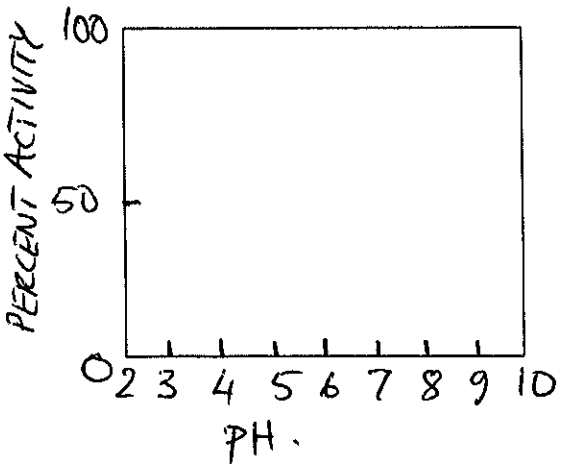


b. Only this protonic form is active

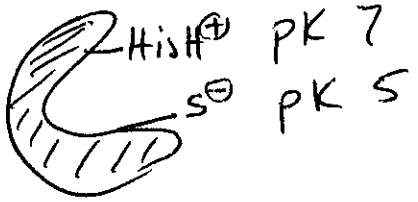




5)



c. Only this protonic form is active

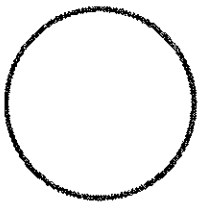


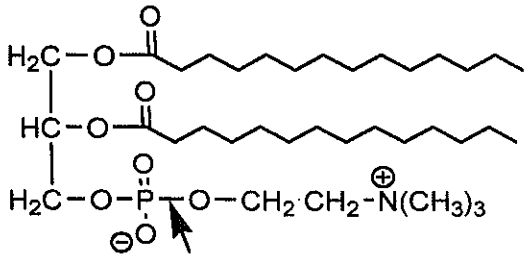
Question 8 (10 pts) Suppose that an enzyme is a tetramer of identical subunits in equilibrium with monomers. The substrate of the enzyme, A, binds 10 times more tightly to the monomer. The monomer is 20-times more active than the tetramer. An allosteric molecule binds preferentially to the tetramer. Monomers react 8-times more rapidly with iodoacetate.

What is the effect of:

Increasing [A] on the proportion of monomers	increase	NC	decrease
Raising the concentration of the allosteric molecule on enzyme activity	increase	NC	decrease
Lowering total enzyme concentration on the percentage of monomer	increase	NC	decrease
Increasing total enzyme concentration on the binding of A	increase	NC	decrease
Increasing the concentration of the allosteric molecule on the reactivity with iodoacetate	increase	NC	decrease

Question 9 (8 pts) vesicles are spherical droplets of buffer enclosed by a continuous lipid bilayer. A cross-section is shown to the right. The bilayer is formed from the phospholipid, phosphatidylcholine, shown at the far right. When this phospholipid is hydrolyzed by phospholipase D the bond shown with an arrow is broken.





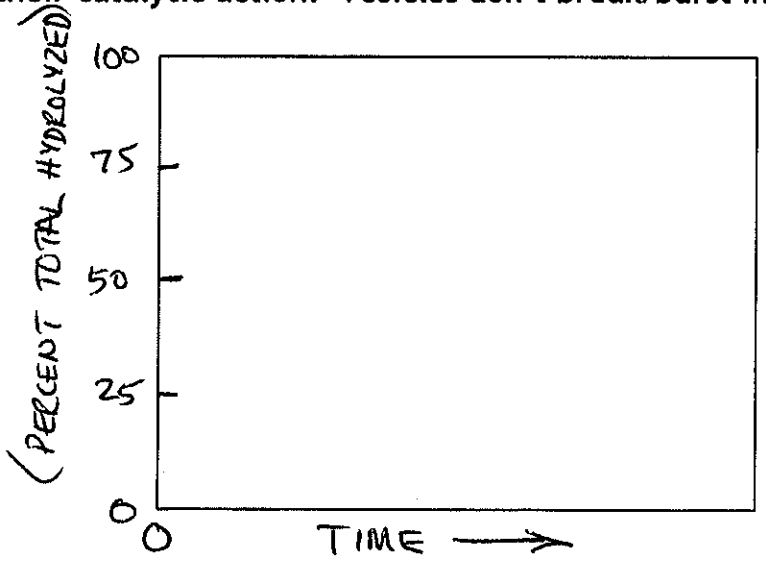
In the spaces provided draw accurate chemical structures of the <u>two products</u> of this reaction.

Suppose that you have 100 vesicles in buffer and then you add 20 enzyme phospholipase D molecules to the buffer. They either

(A) bind to the surface of the first vesicle they encounter, catalyze the reaction as above, but never let go of the vesicle.

Or (B) hop on and off vesicles during their catalytic action. Vesicles don't break/burst in either A or B.

Pay careful attention to the axes of the graph (percentage of the total phospholipid hydrolyzed versus time) and plot the release of choline with time after the addition of enzyme at time zero.



7 20

Question 10 (12 pts.) Fill in the blanks with not more than 3 legible words.

a. Name an irreversible inhibitor of an enzyme _____

b. and the enzyme that is the target of your answer in "a" _____

c. give the NAME of an enzyme that hydrolyzes alpha 1-6 glycosidic bonds _____

d. What compounds can be used to increase the viscosity of solutions in enzymology _____

e. these enzymes do not follow Michaelis Menten Kinetics _____

f. Name a saponifiable lipid _____

g. a technique used to measure lateral diffusion in membranes _____

h. this particular polysaccharide forms a blue color with iodine _____

i. the T state is less soluble in this hemoglobin mutation _____

j. these inhibitors cannot be dialyzed away from the enzyme they inactivate _____

k. these inhibitors change Km not Vmax _____

zz. the word that best describes this exam _____

Life is a struggle with equilibrium that we all eventually lose