

Name _____

Perm _____

1. For the following hypothetical metabolic pathway:

	A → B	B → C	C → D	D → E	E → F	F → G	G → H	H → I	I → J	J → K	K → L	L → M	M → N	N → O	O → P	P → Q	Q → R	R → S	S → T	T → U	U → V	V → W	W → X	X → Y	Y → Z	Z → A
A → B	0.3	1.1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
B → C	0.1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
C → D	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
D → E	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
E → F	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
F → G	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
G → H	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
H → I	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
I → J	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
J → K	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
K → L	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
L → M	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
M → N	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
N → O	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
O → P	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
P → Q	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
Q → R	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
R → S	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
S → T	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
T → U	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
U → V	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
V → W	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
W → X	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
X → Y	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
Y → Z	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		
Z → A	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		

MCDB 108B - Biochemistry

Winter 2012

Answer the following questions. Do not use an equation. Show all your work. When applicable, a line graph with log scale is encouraged.

Midterm

Answer Key.

February 8, 2012

Make sure your exam contains pages numbered 1 through 5. Answer all questions. The point allocation for each question is approximate.

1.4 kcal/mol = 10-fold displacement away from equilibrium (37°C)

Question	
1	_____
2	_____
3	_____
4	_____
total	/67

1. For the following hypothetical metabolic pathway:

	ΔG^0	ΔG
$A \leftrightarrow B$	-0.3	1.1
$B \leftrightarrow C$	0	-5.6
$C \leftrightarrow D$	-1.1	-1.1
$D \leftrightarrow E$	5.6	-1.4
$E \leftrightarrow F$	-1.8	0.6
$F \leftrightarrow G$	1.4	-0.4
$G \leftrightarrow H$	-7.0	0
$H \leftrightarrow I$	0.7	0.2
$I \leftrightarrow J$	0.4	-1.5
$J \leftrightarrow K$	-0.5	-0.3

Answer the following questions. Do not use an equation. Show all your work. When applicable, a line graph with log scale is encouraged.

a) Indicate the step(s) that are candidate points for metabolic control. Explain.(2)

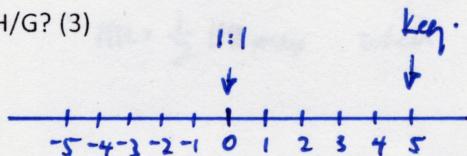
$B \rightarrow C$ ΔG is large & neg. (ΔG^0 is not relevant) \therefore This is the rate limiting step.

b) What is the steady-state molar ratio of D/C? (2)

$\Delta G^0 = \Delta G$ ΔG^0 always implies initial conditions of 1:1 D:C.
 \therefore Initial s.s. D:C is 1:1.

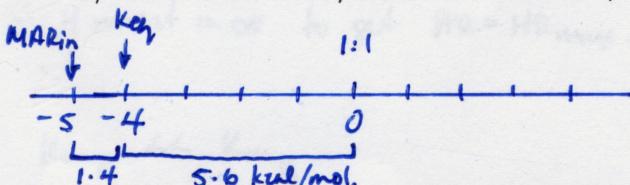
c) What is the steady-state molar ratio of H/G? (3)

$$\frac{7.0}{1.4} = 5 \text{ orders of mag.}$$



$K_{eq} = 10^5$ $\Delta G = 0$ so during s.s. we are at equil. $\therefore M A R_{init} = \underline{\underline{10^5}}$

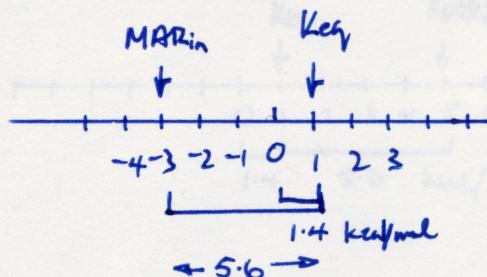
d) What is the steady-state molar ratio of E/D? (4)



$$M A R_{init} = \underline{\underline{10^{-5}}}.$$

e) What is the steady-state molar ratio of D/A? (5)

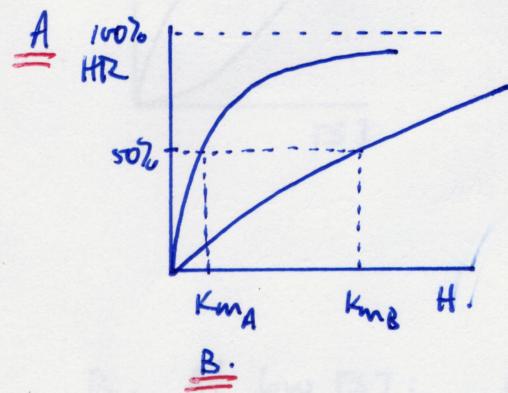
ΔG^0	ΔG
-0.3	1.1
0	-5.6
-1.1	-1.1
-1.4	-5.6



$$M A R_{init} = \underline{\underline{10^{-3}}}$$

2. A hormone at low concentration binds predominantly to a cell surface "receptor A" but not "receptor B", which elicits a particular cellular response. The relationship between bound receptor and free hormone concentration is given by: $HR = HR_{max} \times H / (H + K_m)$, where HR is the amount of hormone-receptor complex at a given hormone concentration. HR_{max} is the maximum amount of HR complex possible. H is the concentration of free hormone. (20)

- a. Draw the expected graph of HR (Y axis) vs. free H (X-axis) for receptor A. Label the axes.
- b. Indicate HR_{max} on the graph, and show how K_m may be determined graphically.
- c. Show mathematically that K_m is the hormone concentration when 50% of the receptors are bound.
- d. What concentration of H results in 100% of the receptors bound? Show this mathematically.
- e. At high hormone concentration, both receptors A and B are significantly activated, causing a different cellular response. Describe how hormone binding to receptors A vs. B differ from each other in terms of their respective kinetic parameters. Show this graphically (you can superimpose on the same graph from a).



C.
$$HR = HR_{max} \times \frac{H}{H + K_m}$$

when $K_m = H$:

$$HR = HR_{max} \times \frac{1}{2}$$

$$\therefore HR = \frac{1}{2} HR_{max} \text{ when } H = K_m.$$

D. If $H = \infty$, $HR = HR_{max} \times 1$.

$\therefore H$ must $= \infty$ to get $HR = HR_{max}$.

E. $K_m_A \ll K_m_B$

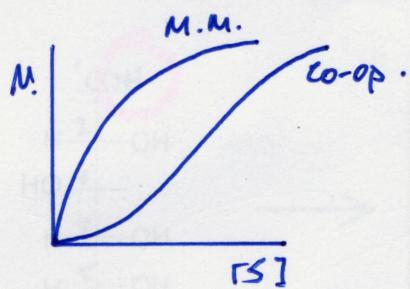
Assume HR_{max} is the same for both.

4. Shown below is a 6-carbon sugar. You wish to design a simple strategy by which this compound can

3a. An enzyme displays cooperative behavior with respect to the substrate dependency of the velocity. What are the essential differences between an enzyme that displays cooperative as opposed to Michaelis-Menten behavior? List them and explain clearly. Draw the graph of velocity vs. substrate concentration for both types of behavior (10). Assume the concerted model.

b. At low substrate concentration the activity of the cooperative enzyme is significantly less than that displayed by the Michaelis-Menten enzyme. But at high substrate concentration, the respective activities are very similar. Explain why. (5) Explain the molecular basis.

c. An allosteric effector is seen to increase the enzyme velocity at any given substrate concentration. In addition, the kinetic behavior becomes more like Michaelis-Menten, while cooperativity is lost. Explain the likely mode of action of this effector. (3)



- A. 1. Multiple binding sites / subunit-complex.
2. T is in equilth with R form.
3. K_m/K_a is very large.
4. S binds to R with much > affinity than to T.

B. At low [S]: $\frac{M \cdot M}{4(R)}$

Co-op
 $\downarrow\uparrow$ 88 R
 $\uparrow\uparrow$

In M.M.: all of the enz is in R form \therefore N is high.

$\uparrow\uparrow$ $\boxed{\text{R}}$ T.

In co-op: most of the enz is in T form \therefore N is very low.

At high [S] ↑ O R

$\uparrow\uparrow$ 88 R

In both M.M. & co-op enz is predominantly in R form \therefore N is high in both.

$\uparrow\uparrow$ $\boxed{\text{T}}$

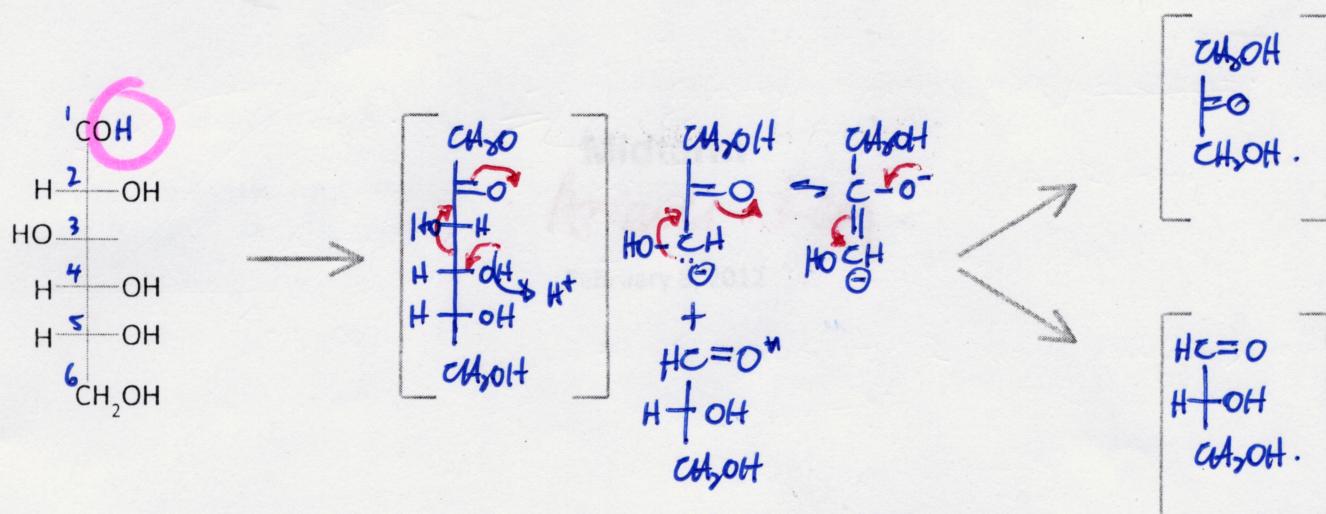
C. An allosteric activator A's N by promoting the R form. The R form displays M.M. behavior \therefore the system becomes more M.M.-like.

4. Shown below is a 6-carbon sugar. You wish to design a simple strategy by which this compound can be cleaved into two 3-carbon compounds.

a. What would be the first reaction step in your scheme? Explain the strategy. Draw the product. (3)

b. Consider the step in which actual cleavage occurs. Show the organic chemical mechanism resulting in cleavage in solution (as opposed to enzymatic). Draw the products. (5)

c. To design an enzyme to more efficiently catalyze the cleavage reaction, what critical amino acid residues would you incorporate into the active site? Explain the strategic role of each residue. (5)



a. Conversion of aldehyde to ketone. Carbonyl grp is needed at C2 to stabilize neg charge that develops during cleavage.

b. see above.

S. 1. Lysine to create Schiff base ($\text{C}=\overset{\oplus}{\text{NH}}-$). Pos. charged quaternary 'N' is a better e^- sink than $\text{C}=\overset{\ominus}{\text{O}}$. Better stabilizes neg charge on C_3 .

2. Tyrosine catalytic base to effectively deprotonate -OH at C₄. Effective conc' of an active site residue is extremely high compared to sol'n concentrations.