MCB102 First Midterm Exam Spring 2003

NAME _____

SID _____

TA _____

1 9 (8 points)

2 6 (7 points)

3 6 (7 points)

4 5 (8 points)

5 8 (8 points)

6 14 (14 points)

7 6 (9 points)

8 9 (9 points)

9 _____ (6 points)

10 _____ (8 points)

11 _____ (7 points)

12 30 (9 points)

Total _____ (100 points)

93

$pH = pK_a + \log [A^-]/[HA]$

$\Delta G'^0 = -2.3\ RT \log K'_{eq}$

$R = 8.3\ J/mol \cdot K$

$$E + S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ES \xrightarrow{k_2} E + P$$

$$E + I \underset{k_{-I}}{\overset{k_I}{\rightleftharpoons}} EI$$

$K_m = (k_{-1} + k_2)/k_1$

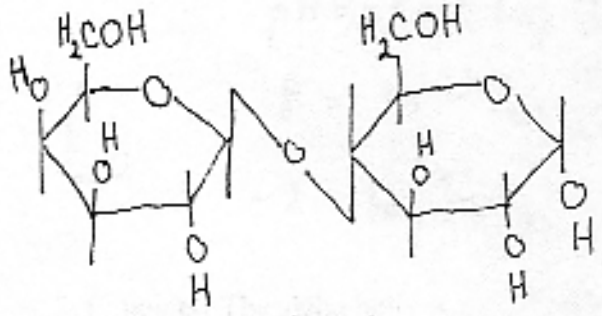
$k_2 = V_{max}/[E_T]$

$K_I = k_{-I}/k_I$

1. Name the structures given below. In the case of amino acids, give the full name or the three-letter abbreviation, as well as the one-letter code. (8 points)

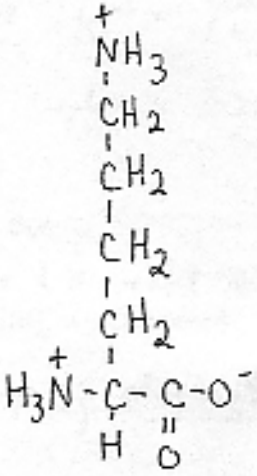
9

(a)



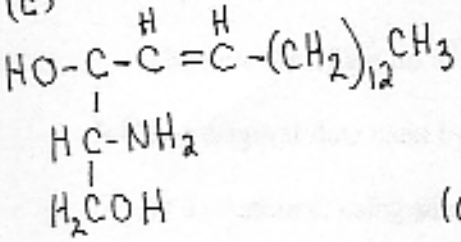
lactose

b.



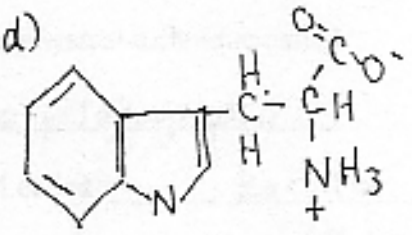
lysine, Lys, K

(c)



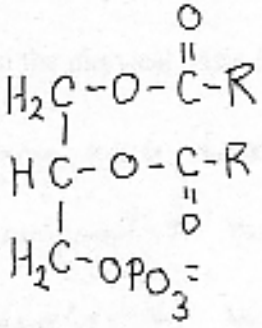
sphingosine

(d)



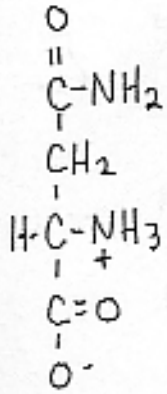
tryptophan, Trp, W

(e)



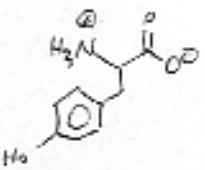
phosphatidic acid

(f)



asparagine, Asn, N

9



6

2. (6 points) The side chain of tyrosine has a pK_a of 10. What fraction of tyrosine molecules have lost their side chain's dissociable hydrogen at pH 8? Show your reasoning.

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

$$8 = 10 + \log \frac{[Tyr^-]}{[Tyr]}$$

$$-2 = \log \frac{[Tyr^-]}{[Tyr]}$$

$$10^{-2} = \frac{[Tyr^-]}{[Tyr]}$$

$$\frac{1}{100} = \frac{[Tyr^-]}{[Tyr]}$$

("The" fraction is $\frac{1}{101}$, I suppose.)

At pH 8, for every 100 undissociated Tyr molecs., there is 1 dissociated Tyr molecule.

6

3. (7 points) The alpha helix is right (1 point)-handed, which means that as you spiral down the helix, you are going clockwise. (2 points)

(looking down)

The number of amino acids per turn is 3.6 (1 point)

6

The original data used by Pauling to propose this structure was obtained by W.T. Astbury, using a physical technique called X-ray crystallography (2 points) and a biological material called keratin (1 point).

(in wool)

fibroin in silk?

4. (8 points) Aliphatic amino acids tend to be clustered in the inside of globular proteins.

5

a. What term describes this phenomenon? (2 points) hydrophobic interactions

b. BRIEFLY explain the physical basis for this phenomenon. (6 points)

-3

Aliphatic amino acids have hydrocarbon R-groups, which are nonpolar, compared to the polar aqueous solutions proteins usually find themselves in. Hence, these nonpolar amino acids tend to be grouped and tucked away inside globular proteins, protected by more hydrophilic portions of the peptide. Since you want this to be brief, I'll just mention that this is also favored because it decreases the entropy of the system.

5. (8 points) A peptide is treated with cyanogen bromide and yields free methionine plus a peptide.

(a) Where is the Met located in the peptide? (1 point) Met / .....

at the amino terminus (the N-terminus)

(b) When treated with trypsin, the original peptide yields the following peptides: R, K at carboxyl end

(4) Thr
(5) AsnLys
(2) GlySerProTrpAlaArg
(1) MetPheLeuLeuArg

When treated with chymotrypsin, the original peptide yields the following peptides: F, W, Y at carboxyl end

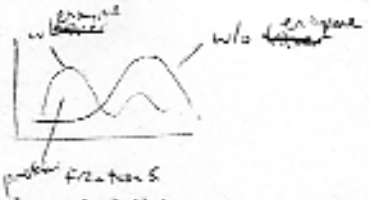
(1) MetPhe
(3) AlaArgAsnLysThr
(4) LeuLeuArgGlySerProTrp

(b) What is the entire sequence of the original peptide? (5 points)

Met Phe Leu Leu Arg Gly Ser Pro Trp Ala Arg Asn Lys Thr

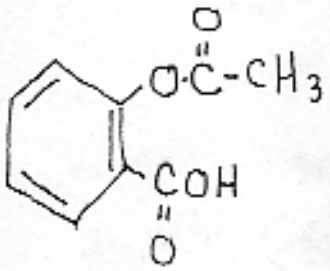
(c) When you analyze the original peptide using the Edman degradation, the first round will yield the amino acid

methionine, which is at the amino end of the peptide. (2 points).



6. (14 points) Aspirin, shown below, inhibits an enzyme involved in complex lipid synthesis. When radioactive aspirin is mixed with this enzyme, and the mixture is passed over a size exclusion column, a substantial fraction of the label comes out of the column early, whereas the remainder emerges later. One hundred percent of the aspirin would emerge later if it had not been mixed with enzyme. All of the protein emerges in the early fractions, and it has no enzyme activity. What kind of inhibition is involved here, and why does some of the labeled aspirin appear in the material that comes off the column early? Be sure to mention how size exclusion chromatography works.

14



Size exclusion chromatography, which involves filling the column with polysaccharide beads with small pores, works by slowing down the smaller proteins in the solution so that they are eluted later.

Irreversible inhibition (specifically, suicide inhibition) is at work here. In suicide inhibition, inhibitors act like the substrate and enter an enzyme's active site. But once the enzyme begins to act upon the inhibitor, it gets stuck, and an enzyme-inhibitor complex is formed - the enzyme can't break free. Thus, in this problem, the aspirin formed a compound with the enzyme, making a large molecule that was able to travel through the column faster than the inhibitor alone, which appeared in later fractions. All the enzymes were inactivated, and hence there was no enzyme activity. This also explains why some labeled aspirin came out early - it was a part of the enzyme-inhibitor complex.

7. (9 points) From the plot of velocity versus substrate concentration shown in the figure, obtain the following parameters. The amount of enzyme in the reaction mixture is 10^{-3} mmol. $= E_t = 10^{-3}\ \text{mmol} \times \frac{10^3\ \mu\text{mol}}{1\ \text{mmol}} = 1\ \mu\text{mol}$

6

The answer to 7a was correct; three points were added.

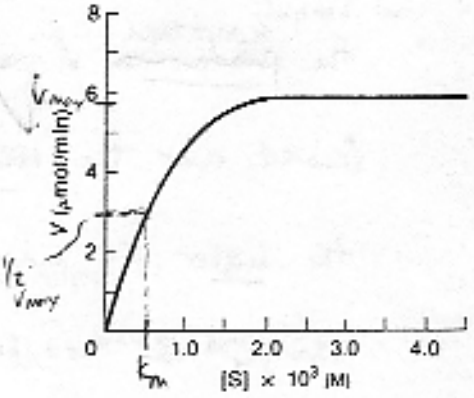
a. (3 points) $K_m \approx 0.5 \times 10^{-3}\ M$ (= 500 M)

b. (2 points) $V_{max} \approx 6\ \mu\text{mol/min}$

c. (4 points) Turnover number

$k_{cat} = V_{max}/E_t$

$= 6\ \frac{\mu\text{mol}}{\text{min}} \cdot \frac{1}{1\ \mu\text{mol}} = 6\ \text{min}^{-1}$



my math is a little slow today...

8. (9 points) A compound with a structure similar to that of proline inhibits both proline racemase and proline hydroxylase. The K_I of this inhibitor for proline racemace is 0.023 micromolar, and the K_I for proline hydroxylase is 0.18 micromolar. Which enzyme is more strongly inhibited? BRIEFLY explain your reasoning.

9

$K_I = \frac{[S][I]}{[SI]}$ (Leave the μM units) $\quad K_I = \frac{k_{-1}}{k_1}$

Thus, a high K_I would indicate that are would be more likely to find the substrate (S) and inhibitor (I) separate than as a complex compound (SI), and conversely, a low K_I indicates that the complex is more prevalent. Hence, the proline-like compound is a stronger inhibitor for the enzyme with a lower K_I - proline racemase.

(0.023 < 0.18)
racemase < hydroxylase

DO NOT GRADE THIS

answer on back of this page →

Answer to #8 (I didn't see the formula on the front)

$K_I = \frac{k_{-1}}{k_1}$ where $E + I \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} E \cdot I$

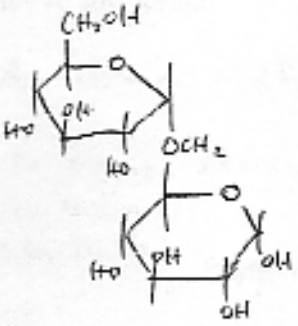
A high K_I indicates that the reverse reaction, that is, the separation of enzyme (E) and inhibitor (I), is favored over the inhibiting reaction. Thus, between two K_I's, the higher K_I indicates the enzyme is being inhibited less strongly than the lower K_I. Hence, in this problem, the proline-like compound is a stronger inhibitor for the enzyme with the lower K_I - proline racemase.

$0.023 < 0.18$

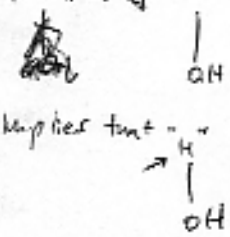
racemase hydroxylase

9 (6 points).

a. (5 points) Draw two molecules of D-glucose in the Haworth convention, joined by an alpha 1→6 glycosidic (full acetal) bond.



all of these ↓



6

c. (1 point) The structure occurs in a biological polymer called

glycogen (which also has α1→4 bonds)

10. (8 points) Consider the reaction S -------> P. Its equilibrium constant is 10,000 when S and P each start at 1 M concentration. What is the standard free energy change? Show your work and include units.

assume T = 298 K

$K'_{eq} = 10{,}000 = \frac{[P]}{[S]}$

8

$\Delta G'^{\circ} = -2.3\ RT \log K'_{eq}$

$= -2.3\ (8.3\ J/K \cdot mol)(298\ K) \log (10{,}000)$

$= -2.3\ (8.3\ J/K \cdot mol)(298\ K)(4)$

$= -9.2\ (8.3\ J/K \cdot mol)(298\ K)$ ← Answer

Answer →

just fooling around

< calculator stuff >

$\Delta G'^{\circ} \approx -30000\ J/mol$

$\approx -30\ kJ/mol$

($-9.2 \cdot 8.3 \cdot 298\ J/mol$)

OK

The standard free energy change is $-(9.2 \times 8.3 \times 298)\ J/mol$.

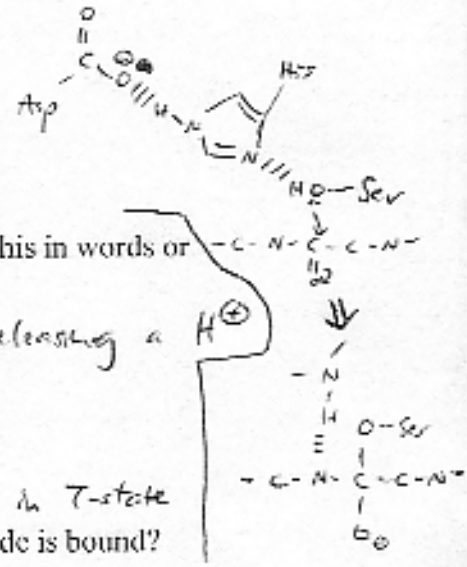
11. (7 points) The serine residue at the active site of chymotrypsin is unusually reactive. BRIEFLY explain the factor(s) responsible for this activity, and explain how these factors facilitate the catalytic role of the active site serine.

The catalytic triad of Asp-His-Ser in chymotrypsin, brought together by the conformation of the enzyme, stabilizes the hydrogen on serine's hydroxyl group (via hydrogen bonding), allowing the oxygen to attack the carboxyl group of the peptide bond in the active site. ~~By allowing the oxygen to partially form a bond with the hydrogen, the~~

7

Water is unable to do this, as there is no easy way to ~~assist~~ deprotonate the oxygen after the ~~carbonyl~~ attack on the carboxyl group.

Figure 1.



12. (9 points) a. How does carbon dioxide bind hemoglobin? Describe this in words or chemical formulae.(5 points)

either ~~d~~directly, forming carbaminohemoglobin and releasing a $H^{\oplus}$

or

indirectly, $CO_2 + H_2O \rightleftharpoons H^{\oplus} + HCO_3^{\ominus}$

↳ binds hemoglobin in T-state

3

b. (2 points) Which state of hemoglobin is stabilized when carbon dioxide is bound?

T-state

2

c. (2 points) What kind of noncovalent bonds between subunits ~~are~~ is stabilized when carbon dioxide is bound?

Certain ionic bonds are stabilized.

2

(the hydrophobic interactions are always there keeping the Hb together)