BIOC *2580

Fall 2012

Practice Package

NB: This package may contain errors that will only be apparent if you come to session.

Instructions: Please attempt all questions prior to the midterm review session.

Megan: Tuesday, October 16, 2012
Library Rm. 384 & 359

These are just additional questions for more practice.
Make sure you do the **problem sets** and **sample midterms** on D2L.

DATA:

Amino Acid Side Chain	pKa
Asp	4.0
Cys	8.5
Arg	12.5
Glu	5.0
Tyr	10.0
His	6.5
Lys	10.0

Average N-terminal: use pKa- 8.0
Average C-terminal: use pKa- 3.0

1. What is the net charge on the following peptide at pH 6.5?

   Glu-Ala-Arg-Met-Gln-Asp-His-Ser

   a. +0.5
   b. +1
   c. -0.5
   d. -1

2. Which chromatography technique separates proteins solely on the basis of size?

   a. Gel Filtration
   b. Reverse Phase
   c. Ion Exchange
   d. Metal Affinity

3. Which peptide would be eluted first in gel filtration chromatography?

   a. Lys-His
   b. Trp-Tyr-Thr
   c. Ala-Leu
   d. Arg-Glu-Gly
   e. His-Thr-Tyr-Ala

4. In a mixture of the four proteins listed below which will elute second in gel filtration chromatography?

   a. Ovalbumin, M=45 kDa
   b. Immunoglobulin, M=145 kDa
   c. Ribonuclease, M=13.7 kDa
   d. Serum albumin, M=68.5 kDa

5. How many fragments are made when the following oligopeptide is cut with chymotrypsin?

   Ala-Phe-Pro-Tyr-Lys-Tyr-Leu-Arg-Lys-Trp-Pro-Trp-His-Ser

   a. 2
   b. 3
   c. 4
   d. 5

6. A poly peptide is cleaved into peptides by treatment with trypsin and cyanogens bromide, and then the peptides are purified and sequenced. The sequences of the peptides are shown below.

Trypsin peptides	Cyanogen Bromide peptides
T-1 GSMDPGDLMTLR	C-1 TLRNAFFGSKFHPQA
T-2 ITGLAIHQK	C-2 ITGLAIHQKELIM
T-3 NAFFGSK	C-3 VPKGSM
T-4 ELIMVPK	C-4 DPGDLM
T-5 FHPQA	

Based on the original sequence on the previous page which of the following polypeptides represents the N-terminus.

   a. T-3
   b. C-1
   c. C-2
   d. T-5

7. The major factor causing a typical protein to fold into a compact globular form is :

   a. Hydrophobic effect and non polar interactions
   b. Ion pair and salt bridges
   c. Disulphide bonds
   d. Hydrogen bonds

8. By what factor does a typical enzyme speed up reaction rates compared to uncatalyzed reactions?

   a. 10,000 fold
   b. 10,000,000 fold
   c. 10,000,000,000 fold
   d. 10,000,000,000,000 fold

9. Which level of protein structure describes all aspects of the three dimensional folding of a polypeptide?

   a. Quaternary structure
   b. Tertiary structure
   c. Secondary structure
   d. Primary structure

10. Megan completed mass spectrometry based sequencing of a peptide from her protein and then accidently lost her peptide sequencing results. She was able to find the mass values for the

peaks of the fragments, listed here in Daltons: 128.09497, 265.15388, 380.18083, 479.24925,578.31767, 707.36026, 870.42359, 957.45562

Using the list of masses given below, what is the identity of the third amino acid in the peptide when the sequence is written from n- to C-terminus?

Amino Acid Masses (kDa)

G: 57.02147	R: 156.10111	K: 128.09497	V: 99.06842
S: 87.03202	H: 137.05891	D: 115.02695	E: 129.04259
T: 101.04768	Y: 163.06333	N: 114.04293	F: 147.06842

a. Thr
b. Asp
c. Val
d. Glu

11. Rotation can happen around which atom?

a. Beta carbon
b. Amino nitrogen
c. Alpha carbon
d. Carbonyl carbon

12. Which of the following statements is not true with respect to enzyme kinetics?

a. KM is equal to the substrate concentration [S] that causes the enzyme to have rate = 0.5 × Vmax
b. KM = 0.5 × Vmax
c. KM is a true constant independent of substrate or enzyme concentration
d. Vmax is only constant if a constant concentration of enzyme is used in each measurement.
e. High KM indicates that a substrate is a poor fit to the enzyme's catalytic site.

13. Which amino acid is most likely to be found on the outside of the protein?

a. Valine
b. Glycine
c. Isoleucine
d. Histidine

14. Edman's method for determining amino acid sequence involves which of the following two distinct steps?

a. Coupling to phenylisothiocyanate in mild base followed by hydrolysis in acid
b. Coupling to phenylisothiocyanate in anhydrous acid followed by cyclization in mild base

c. Coupling to phenylisothiocyanate in mild base followed by cyclization in anhydrous acid
d. Coupling to fluorodinitrobenzene in mild base followed by hydrolysis in acid
e. Coupling to fluorodinitrobenzene in mild acid followed by cyclization in anhydrous base

15. Mixed inhibition including non-competitive can be recognised on a Lineweaver-Burk (double reciprocal) plot which shows that the inhibitor produces

a. Change in slope and V_{max}
b. No change in slope
c. No change in V_{max}
d. No change in initial velocity
e. Change in initial velocity

16. What initial reaction velocity (vo) is observed if substrate concentration in an enzyme reaction is 0.5 × KM and Vmax is 2.4×10^{-6} mol L-1 min-1.

a. 1.2 × 10-6 mol L-1 min-1.
b. 6.0 × 10-7 mol L-1 min-1.
c. 2.4 × 10-6 mol L-1 min-1.
d. 8.0 × 10-7 mol L-1 min-1.
e. 1.6 × 10-6 mol L-1 min-1.

17. What substrate concentration, expressed as a multiple of KM, must be present when an enzyme reaction is observed to have an initial rate vo = 0.75 Vmax.?

a. [S] = 0.25 × KM
b. [S] = 0.75 × KM
c. [S] = 1.33 × KM
d. [S] = 3.0 × KM
e. [S] = 4.0 × KM

18. The absorbance of a solution of NADH is 0.465, measured at 340 nm in a 1.000 cm thick sample. If the extinction coefficient □□= 6200 L mol^{-1}□$dm^{-1}$for NADH, then the concentration of NADH is:

a. 3.47×10^{-4}M
b. 1.37×10^{-3}M
c. 1.33×10^{-4}M
d. 7.50×10^{-5}M
e. 2.55×10^{-2}M

19. Which of the following is true? A protein that has been denatured by urea followed by treatment with Mercaptoethanol can be correctly refolded upon . . .

    a. Removal of urea and mercaptoethanol
    b. Removal of urea and exposure to O_2
    c. Exposure to O_2 followed by removal of urea
    d. Removal of mercaptoethanol and exposure to O_2
    e. Once it has been denatured it cannot be refolded to the native state again

20. Which of the following amino acids is a breaker of regular secondary structure?

    a. Trp
    b. Met
    c. Gly
    d. Val
    e. Phe

21. Which of the following amino acid can accept H-bonds on its side chain?

    a. Phe
    b. Ala
    c. Glu
    d. Leu
    e. Gly

22. Which of the following will most likely have alpha helical secondary structures?

    a. Ala-Leu-Met-Phe-Glu-Gly
    b. Tyr-Trp-Phe-Met-Pro-Gly
    c. Gly-Pro-Asn-Asp-Pro
    d. Ile-Tyr-Val-Thr-Ile
    e. Ala-Ile-Trp-Phe-Val

23. Which amino acid gives a yellow colour when stained with ninhydrin?

    a. Thr
    b. Pro
    c. Tyr
    d. Lys
    e. N-terminal amino acid

24. What is the average net charge of tyrosine at pH 9.6?

a. + 0.72
b. +0.28
c. 0
d. –1.28
e. –0.72

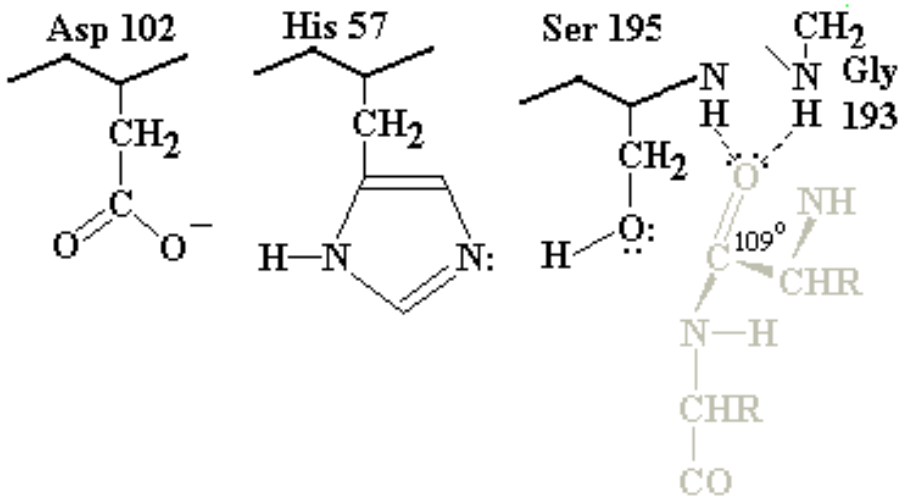
25. Which peptide would be eluted first in cation exchange chromatography at pH 7?

a. Asp- Ala-Glu
b. Tyr-Lys-Pro
c. Leu-His-Phe
d. Tyr-Lys-Arg-His

Short Answer Questions!!!

1. Draw the complete structure of the tri peptide Lys-Pro-Tyr, show all the ionizable groups at a pH of 7, show the net charge of the tripeptide:

2. This question relates to the catalytic triad!



   a. Which amino acid acts as the nucleophile?

   b. Circle the oxyanion hole, what is its main purpose?

   c. What is the role of histidine at the stage of the reaction?

   d. What is the role of the Asp 102?

e. Which amino acids does chymotryspin cut after?

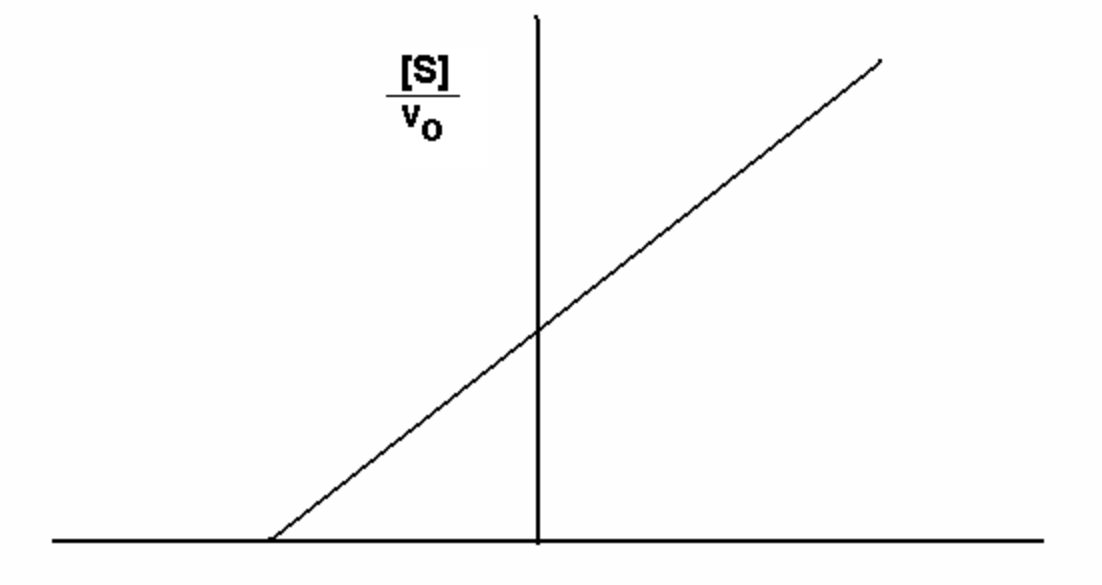
f. Draw in the curly arrows representing where electron pairs go in this step of the reaction.

3. Derive the Michaelis-Menton Equation:

4. Draw a box around each cluster of secondary structure breakers in this polypeptide. Then with the fragments predict the secondary structure.

ITWYHCVTIYFSWDGFQELMRKHHLERQALMPNCRTWEMIHKVLIFF

5. Given the following information, calculate V_{max} and V_o (when [S]=0.22mol/L).



x- intercept = 4.25×10^4 mol L^{-1}
y-intercept = 6.25×10^5 min
slope = 20 L min mol^{-1}

6. The following questions relate to primary and secondary protein structure.

    a. There are three types of secondary structures. What are they?

    b. Why do non-polar amino acids tend to face the interior of the protein?

c. What is the purpose of secondary structure breakers? Name 2!

d. Identify the types of structures shown in the images below.