Practice questions and explanations

BIOS 1006

Week 1

Important things to remember

*that have applications and are not just pure memorization

- As pH increases, compounds become deprotonated (lose an H+).
- Always start with full protonation when finding the pI of a peptide.
- Amino acid properties are determined by the properties of their side chains (i.e. know the properties of functional groups!).

Lecture 1: Fundamentals

Functional groups

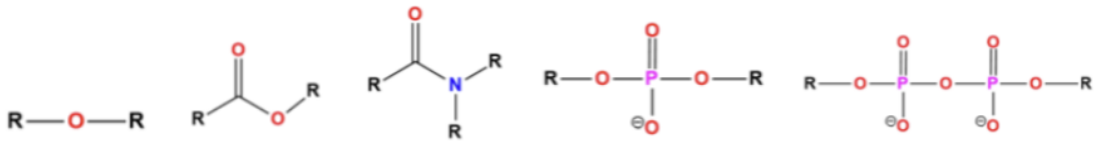
Question 1: Naming

Name the following functional groups.

Answers:

alkyl, hydroxyl, thiol (sulfhydryl), carbonyl, carboxyl, amino, phosphate

Name the following linkages.



Answers:

ether, ester, amide, phosphodiester, phosphoanhydride

Question 2: Roles of functional groups

1. Amino acids and proteins have amino groups & carboxyl groups.
2. Carbohydrates tend to have an abundance of hydroxyl groups and ether linkages.
3. Lipids vary greatly in structure, but fatty acids typically have alkyl groups.
4. Each nucleotide in a nucleic acid molecule has phosphodiester linkages.

pH, pKa, pI

Question 3: ICE tables and Henderson-Hasselbalch equation

You make a 0.2 M aqueous solution of propionic acid CH_3CH_2COOH by dissolving an appropriate amount of propionic acid in water. The pH of the resulting solution is 4.88. What is the pKa of propionic acid?

$$CH_3COOH \rightleftharpoons CH_3COO^- + H^+$$

Set up the ICE table:

	$[CH_3COOH]$	$[CH_3COO-]$	$[H^+]$
Initial	0.2	O	O
Change	-x	+x	+x
Equilibrium	0.2-x	+x	+x

Given that the concentration of H^+ is equal to x, we can undo the logarithm to find $[H^+]$:

$$\begin{aligned} pH &= -\log[H^+] \\ 4.88 &= -\log[H^+] \\ [H^+] &= 1.32 \times 10^{-5}\ M \end{aligned}$$

Now plug this into the Henderson-Hasselbalch equation:

$$4.88 = pK_a + \log\left(\frac{1.32 \times 10^{-5}}{0.2 - 1.32 \times 10^{-5}}\right)$$
$$4.88 = pK_a + \log(6.6 \times 10^{-5})$$
$$4.88 = pK_a - 4.18$$
$$pK_a = 4.88 + 4.18$$
$$pK_a = \boxed{9.06}$$

Question 4: Calculating pI of a peptide

A tripeptide formed from tyrosine, valine, and glycine, in the order as stated. Use the following pKa values: α-COOH = 2.2, α-NH_3^+ = 9.4, tyrosine side chain OH = 10.5.

(a) Draw the tripeptide and clearly label where the N-terminus and C-terminus are at pH 7.

$$\text{(N-term) } NH_3^+ - \text{Tyr} - \text{Val} - \text{Gly} - \text{COOH (C-term)}$$

(b) Calculate the charge of this tripeptide at pH 7.

Compare pKa and pH of each ionizable group:

N-term pKa 9.4 > pH 7 → protonated, charge = +1

C-term pKa 2.2 < pH 7 → deprotonated, charge = -1

Tyrosine side chain pKa 10.5 > pH 7 → protonated, charge = 0 (protonated form, -OH, carries neutral charge)

Overall charge: 0

(c) Calculate the pI of this tripeptide at pH 7.

Order all the pKa values:

C-term 2.2
N-term 9.4
Tyr 10.5

Always assume full protonation at the beginning (pH < pKa). When the molecule is fully protonated, the overall charge is +1 (neutral C-terminus and tyrosine, positive N-terminus). As the pH increases, the C-term deprotonates first (pKa 2.2), then the N-term (pKa 9.4), and finally the side chain of tyrosine (pKa 10.5). The pI is the average of the two pKa values that surround the zero charge state.

+1
C-term 2.2
0
N-term 9.4
-1
Tyr 10.5
-2

The flanking pKas at charge 0 are 2.2 and 9.4.

$$\frac{2.2 + 9.4}{2} = 5.8$$

The pKa at pH 7 of the peptide is $\boxed{5.8}$.

(d) Will you be able to use UV light absorbance at 280 nm to detect your tripeptide? Why or why not?

$\boxed{\text{Yes, because the tripeptide contains a tyrosine side chain}}$, which has a phenolic ring that absorbs UV light at 280 nm. The other two amino acids (valine and glycine) do not absorb UV light at this wavelength.

Lecture 2: Amino acids and peptides

Question 5: Amino acid chemical properties

Which amino acid side chains are ionizable? YECDHKR

Which amino acid has no chiral center? G

Which amino acids have hydrophobic (nonpolar) side chains? AVLIGMPFWY

Which amino acid side chain has an amino group? K

Which amino acids have basic functional groups? RHK

Lecture 3: Protein structure

sadly i can't find anything worth putting here BUT KNOW DIHEDRAL ANGLES!!!

Lecture 4: Lab techniques

Question 6: Protein purification

You are attempting to purify the protein a-antitrypsin from a crude cell lysate in which the main contaminants are the other four proteins, B-E, in the table below. The proteins have the molecular weights (MW) and isoelectric points (pI) values shown in the table. Each of the five proteins contains just a single polypeptide chain.

Protein	**Molecular weight**	**pI**
A. α-antitrypsin	45,000	3.4
B. cytochrome c	13,400	10.6
C. myoglobin	17,000	7.0
D. serum albumen	69,000	4.8
E. transferrin	90,000	5.9

(a) Which protein (A to E) will bind to cation-exchange resin at pH 7.4?

B, because pH $<$ pI, so the molecule is positively charged.

(b) Which protein (A to E) would elute first in size-exclusion chromatography?

E, because it has the highest molecular weight.