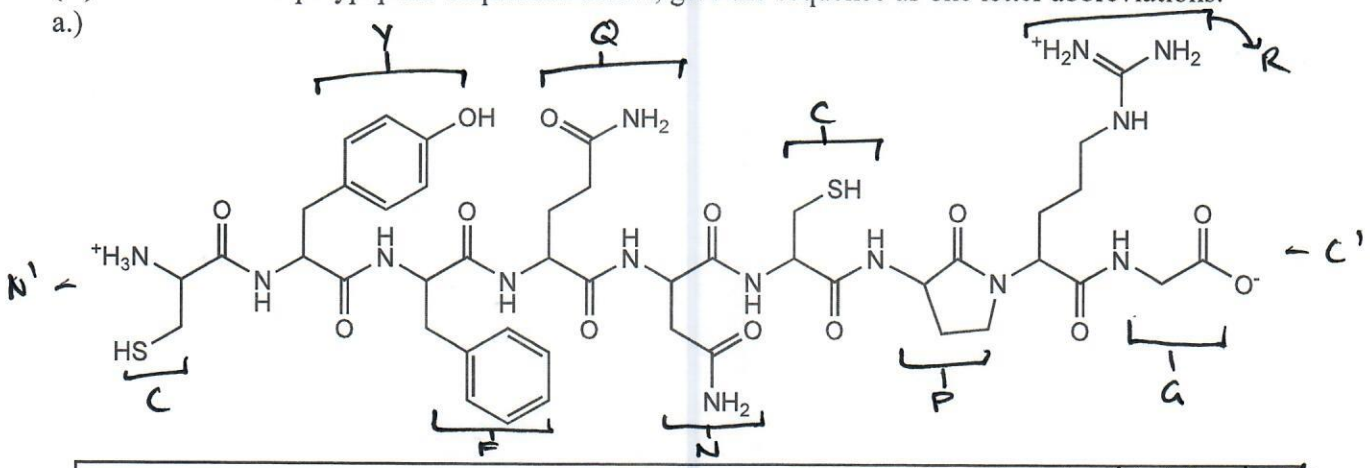
BCHM 463 GSS - 09/12/2022

Acid Base Chemistry, Amino Acids, Protein Structure

(1.) For each of the polypeptide sequences below, give the sequence as one letter abbreviations.

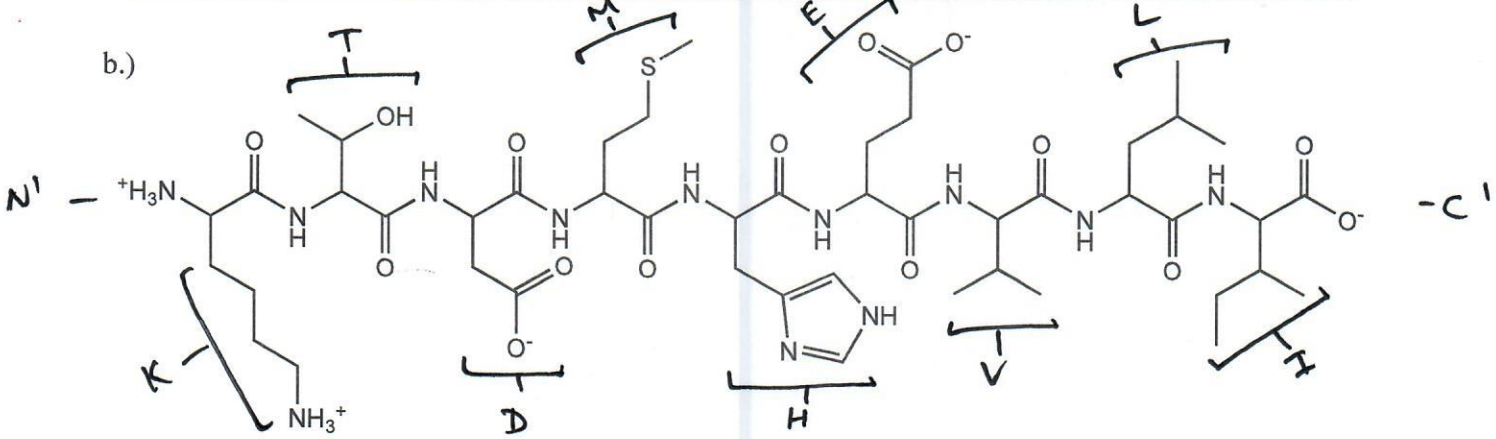
a.)



N'-CYFQNCPRG-C'

* Interesting note - this is the polypeptide sequence for vasopressin hormone.

b.)



N'-KTDMHEVLI-C'

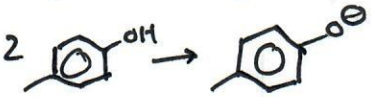
c.) The charge of which peptide would be most affected by a shift in pH from 7.4 (as drawn) to a pH of 12? Explain.

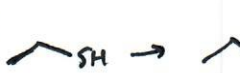
Functional Group	pKa
Carboxyl-terminus	3.5
Amino-terminus	8.5
α-Carboxyl (free amino acid)	2
α-Amino (free amino acid)	9.5
Aspartate R group	3.9
Glutamate R group	4.2
Histidine R group	6
Cysteine R group	8.2
Tyrosine R group	10.0
Lysine R group	10.5
Arginine R group	12.5

Peptide a.)

7.4 → 12 :

Residue 1 

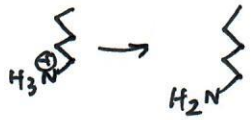
Residue 2 

Residue 6 

Total charge of -3

∴ Peptide a would be the most affected by a shift from 7.4 to 12.

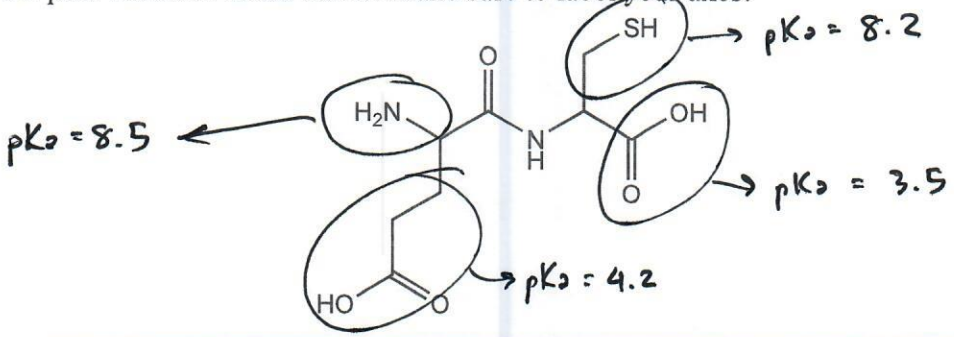
Peptide b.)

Residue 1 

Total charge of -1

Residue

(2.) Below is a structure for the dipeptide Glu-Cys. Draw a titration curve for this dipeptide, then determine the pI, given the pKa values as listed below. Make sure to label your axes!



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Aspartate R group	3.9
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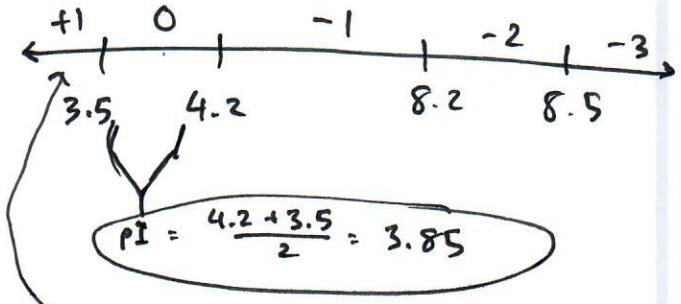
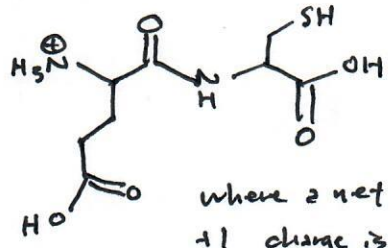
Histidine R group	6
Cysteine R group	8.2
Tyrosine R group	10.0
Lysine R group	10.5
Arginine R group	12.5

(Not to scale)

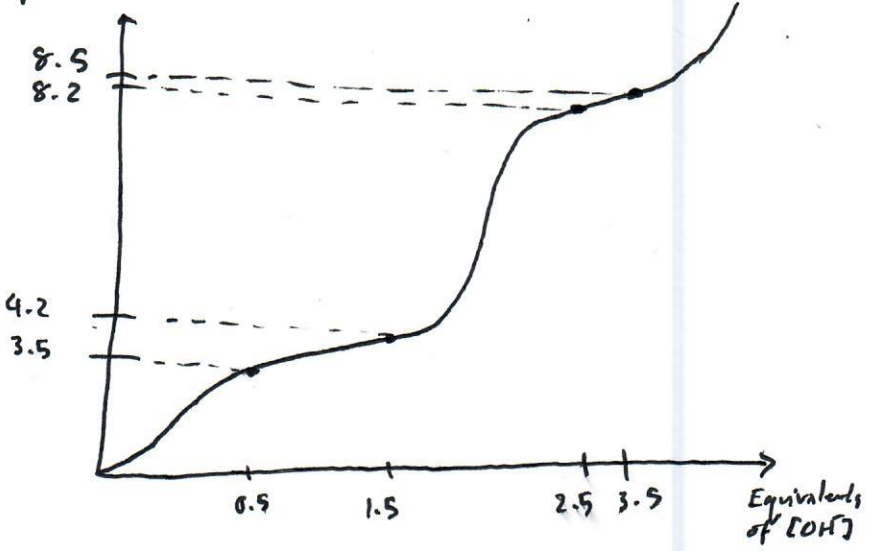


At pH = 3 for instance, ~~molecule~~ Glu-Cys would exist in the form:

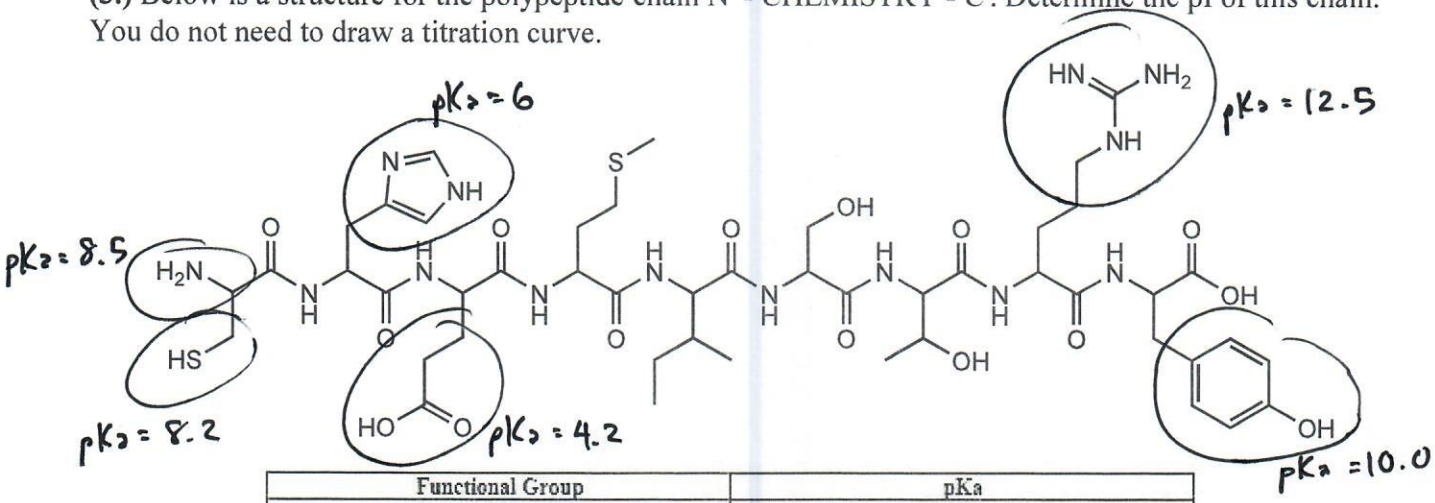


where a net +1 charge is present.

(Graph not to scale)



(3.) Below is a structure for the polypeptide chain N' - CHEMISTRY - C'. Determine the pI of this chain. You do not need to draw a titration curve.



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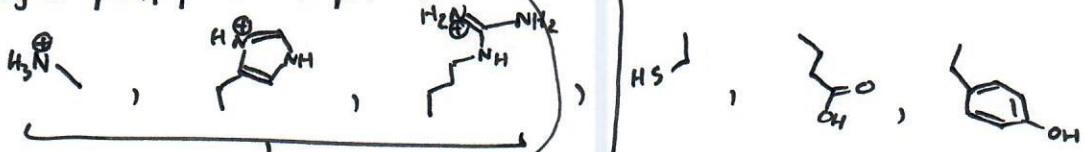
Histidine R group	6
Cysteine R group	8.2
Tyrosine R group	10.0
Lysine R group	10.5
Arginine R group	12.5

No repeat residues, increment by −1 with increasing pH.

① +3 +2 +1 0 −1 −2 −3

4.2 6 8.2 8.5 10.0 12.5

② Imagine polypeptide at pH = 3.0 :



+3 → at pH = 3

$$pI = \frac{8.2 + 8.5}{2} = 8.35$$

(4.) Below is a table of common biological buffers and their pKa values.

a.) In the research lab you are working at, you are asked to make 1L of buffer for a system at a pH of 8.0. You have several options for making this buffered system, as detailed below. Which of the choices would be the best? Explain why the other mixtures would not be optimal.

Choice	Buffer Material	pKa	Moles Conjugate Acid	Moles Conjugate Base
A	$H_2CO_3 \rightleftharpoons HCO_3^{-2}$	6.77	9.5	2.5
B	Tris	8.1	10	0.5
C	MOPS	7.2	1.2	8.35
D	Glycylglycine	8.27	0.1	2

Use $pH = pK_a + \log_{10}\left(\frac{[A^-]}{[HA]}\right)$ Buffer is good when solution is ±1 of desired pH. ↳ in this case, (7.0, 9.0)

Thus,

(A) $pH = 6.77 + \log_{10}\left(\frac{2.5}{9.5}\right) = 6.190$ ✗

(B) $pH = 8.1 + \log_{10}\left(\frac{0.5}{10}\right) = 6.799$ ✗ } Too low

(C) $pH = 7.2 + \log_{10}\left(\frac{8.35}{1.2}\right) = 8.043$ ✓ → within range

(D) $pH = 8.27 + \log_{10}\left(\frac{2}{0.1}\right) = 9.571$ ✗ → Too high

MOPS buffer with 1.2 Moles of MOPS-H^+ and 8.35 Moles of MOPS is the best choice.

b.) Suppose you need to make three different solutions of MOPS buffer that each contain a total MOPS concentration (acid + conjugate base) of 0.5 M but are at different pH values. For each of the following pH values, what are the concentrations of the acid form of MOPS and the conjugate base form of MOPS?

pK_a of MOPS = 7.2

↳ Need $[A^-]$ and $[HA]$:

Rearrange $pH = pK_a + \log_{10}\left(\frac{[A^-]}{[HA]}\right)$

$10^{(pH - pK_a)}[HA] = [A^-]$

Since total [MOPS] = 0.5 M,

$[HA] + [A^-] = 0.5$,

$[A^-] = 0.5 - [HA]$

$10^{(pH-pK_a)}[HA] = 0.5 - [HA]$

$\left(10^{(pH-pK_a)} + 1\right)[HA] = 0.5$

i.) pH = 5

$\left(10^{(pH-pK_a)} + 1\right)([HA]) = 0.5$

$1.0063\,[HA] = 0.5$

$[HA] = 0.4969\,M$, $\therefore [A^-] = 0.00314\,M$

ii.) pH = 7

$\left(10^{7-7.2} + 1\right)[HA] = 0.5$

$[HA] = 0.306286\,M$, $\therefore [A^-] = 0.1937$

iii.) pH = 10.5

$\left(10^{10.5-7.2} + 1\right)[HA] = 0.5$

$[HA] = 2.505 \times 10^{-4}$, $\therefore [A^-] = 0.4997$

c.) For the buffer chosen in part (a), would the buffer solution be more resistant to decreases or increases in the pH of the system? Explain why.

The buffer should be more resistant to decreases in the pH of the system. There are 8.35 moles of MOPS conj. base to counteract the addition of any acids, but only 1.2 moles of MOPS conj. acid to counteract the addition of bases.

(5.) Suppose the polypeptide chain N' - GLSCRIQDEFECY - C' spontaneously forms an α-helix in a solution with a pH of 7.4.

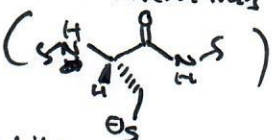
a.) Name at least 3 *different* intrahelical interactions between side chains in the polypeptide chain. Make sure to specify what amino acids are interacting, and what kind of interaction is present.

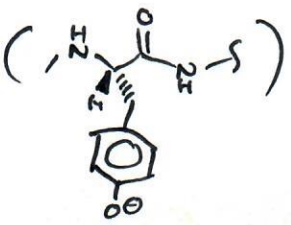
There are many answers, but here are a few:

Induced Dipole	H-Bonding	Ionic Interaction
L2 - I6	S3 - Q7	R5 - D8 R5 - D9

b.) If the pH is increased from 7.4 to 11, the secondary α–helical structure of this chain becomes distorted. Why might this be happening? Consider how different pH environments may affect intrahelical interactions of the polypeptide chain.

Functional Group	pKa
Carboxyl-terminus	3.5
Amino-terminus	8.5
α-Carboxyl (free amino acid)	2
α-Amino (free amino acid)	9.5
Aspartate R group	3.9
Glutamate R group	4.2
Histidine R group	6
Cysteine R group	8.2
Tyrosine R group	10.0
Lysine R group	10.5
Arginine R group	12.5

Most likely the more basic environment is introducing more negative charges that may create unfavorable side chain interactions. For example, at pH = 11, C12 is deprotonated () which may clash with the already negatively charged D8 (Asp) and E9 (Glu) residues to destabilize the α-helix.

Similarly, Y13 is deprotonated at a pH of 11 () which would repel the E9 side chain as well. C4 would also repel D8.