CONCEPT: ISOELECTRIC POINT OF A PEPTIDE

●Recall: pI is always the midpoint between the _____ pK's for the _____ ionizations involving the *neutral* species.

□ pI of a peptide can only be _____ because of the *unique* microenvironment.

□ True pI values for peptides must be _____ determined.

□ Recall: Be sure to use pK_a values for amino acid _____, *not* pK_a's for *free* amino acids.

●For pI of a peptide, follow similar steps for calculating pI of amino acids with *ionizable R-groups*.

Step #1: _____ how Amino Acid R-groups ionize.

Step #2: _____ the pK_a values from smallest to greatest.

$pK_{a1} < pK_{a2} < pK_{a3} < pK_{a4}$

Step #3: Determine net _____ of predominate structures at any pH between each pair of adjacent pK_a's (by comparing pH to all pK_a values).

Net Charge:

pH: pK_{a1} pK_{a2} pK_{a3} pK_{a4}

Structure #1 Structure #2 Structure #3

Imagine/draw predominate structures to determine their net charges.

Step #4: Calculate pI by _____ the two pK_a values "sandwiching" the pH where the predominate structure has a *neutral* net charge.

$$pI = \frac{pK_a + pK_a}{2}$$

EXAMPLE: Estimate the isoelectric point for the following peptide: D-G-E.

a) 3.7

b) 4.5

c) 6.2

d) 11.9

Ionizable Group		pK_a
C-terminus	—COOH	3.5
Asp	—CH_2—COOH	3.9
Glu	—CH_2—CH_2—COOH	4.1
His	—CH_2—imidazole	6.0
Cys	—CH_2—SH	8.4
N-terminus	—NH_3^+	8.0
Tyr	—CH_2—phenol	10.5
Lys	—$(CH_2)_4$—NH_3^+	10.5
Arg	—$(CH_2)_3$—NH—C(=NH_2^+)—NH_2	12.5

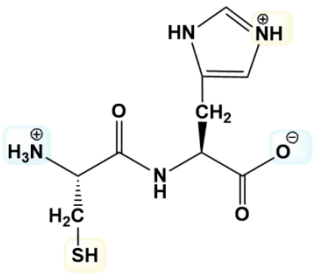
PRACTICE: Estimate the isoelectric point of the following dipeptide in the figure.

a) 4.75

b) 7

c) 9.5

d) 10.5



PRACTICE: Calculate the approximate pI of the peptide: C-G-E-K.

a) 3.08 b) 5.71 c) 6.05 d) 9.63

PRACTICE: Draw in the R-groups for the following peptide & calculate the pI: ATLDAG.

a) 7.41

b) 6.9

c) 9.25

d) 3.7