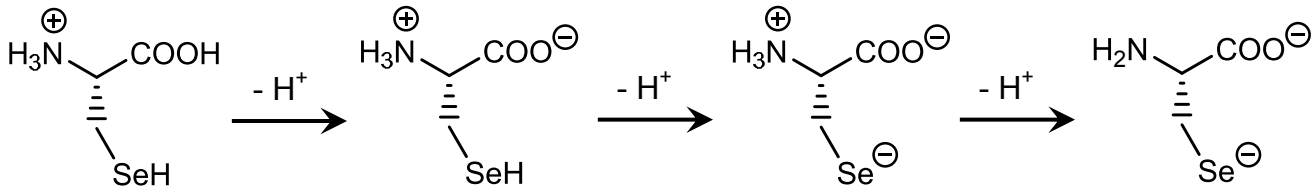
Chemistry 2302

Workshop 25 Solutions
Analyzing Non-Standard Amino Acids

1. To find the pI of an amino acid, we need to determine the pH range over which it is neutral. The pI is in the middle of this range. I usually do this by drawing all of the protonation states of the amino acid, starting with the fully protonated state, and then successively remove protons with increasing pH.

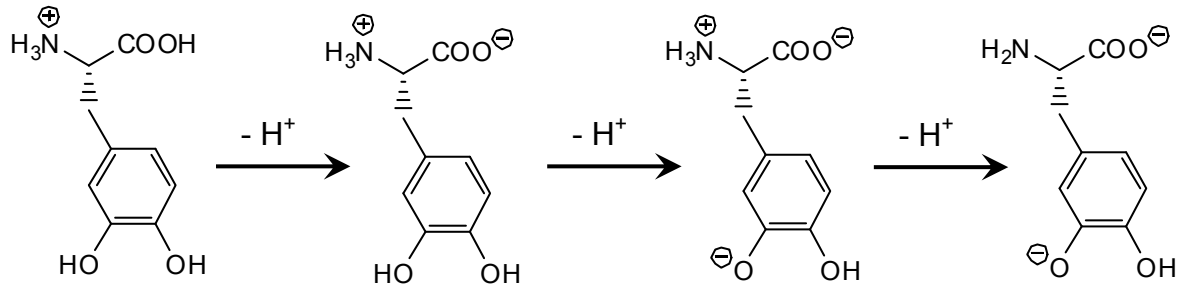
   Selenocysteine has three acidic groups: a carboxylic acid (pK_a ≈ 2.5), the selenol group (pK_a ≈ 5.2), and an ammonium cation (pK_a ≈ 9.5).



pH < 2.5	2.5 < pH < 5.2	5.2 < pH < 9.5	pH > 9.5
(charge = +1)	(charge = 0)	(charge = -1)	(charge = -2)

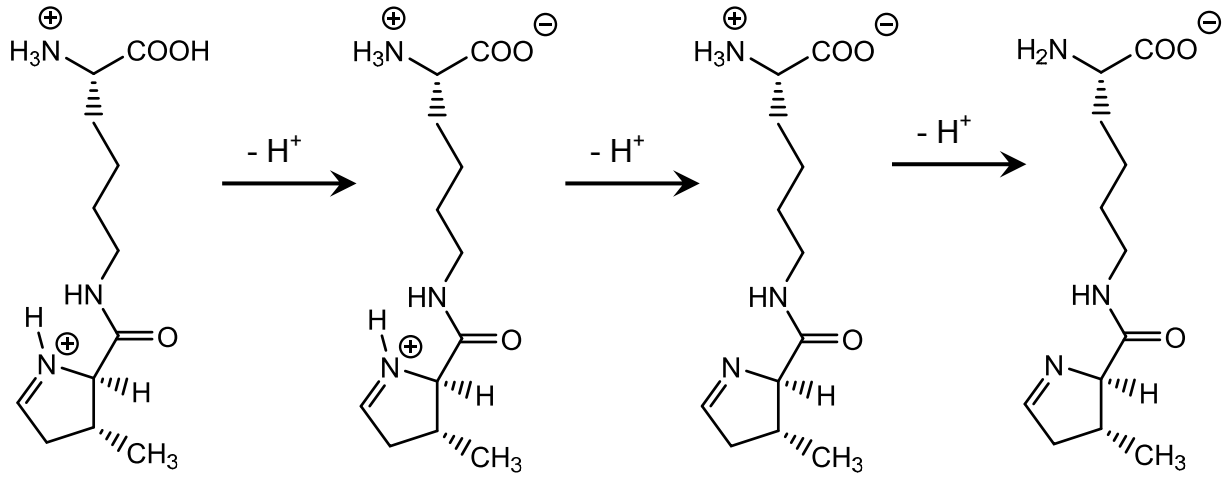
   We can calculate pI of any polyprotic molecule (a molecule with multiple acidic/basic groups) by averaging the pK_a's of the acid-base events on either side of the charge = 0 state. Here, that's the average of 2.5 and 5.2, or **pI = 3.9**.

   Fully protonated L-DOPA has three acidic groups—the carboxylic acid and ammonium cation present in all amino acids, and a catechol (dihydroxybenzene). (The catechol would probably deprotonate only once, at either -OH, to form a hydrogen-bonded anion; the cost of deprotonating twice, and of having two anions right next to each other, would probably insure just one deprotonation.)



pH < 2.5	2.5 < pH < 9.0	9.0 < pH < 9.5	pH > 9.5
(charge = +1)	(charge = 0)	(charge = -1)	(charge = -2)

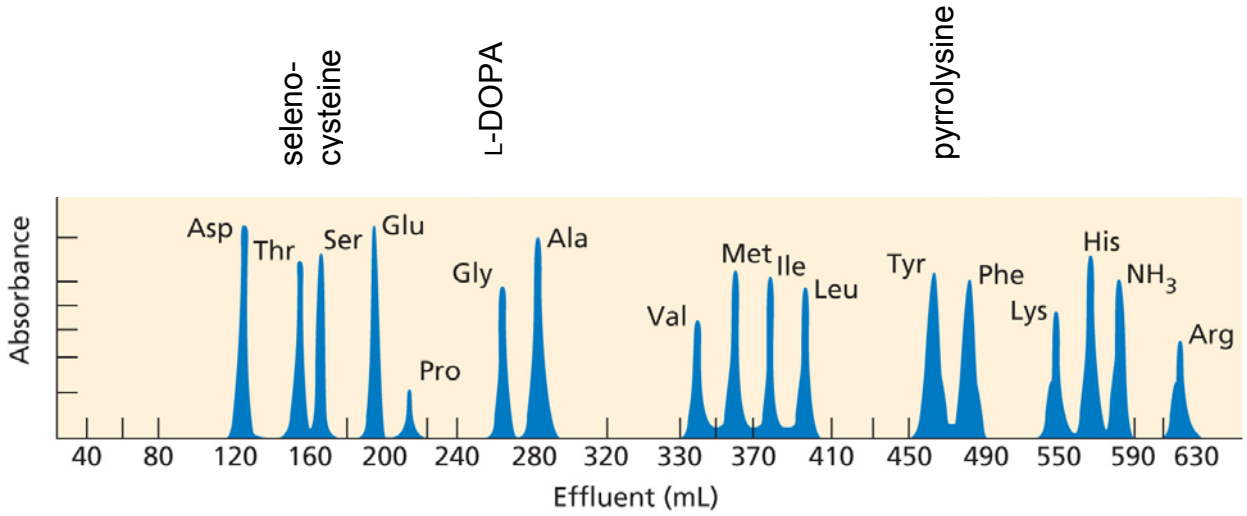
Once again, the isoelectric point will be the average of the pK_a's of the acid-base events on either side of the charge = 0 state. Here, that's the average of 2.5 and 9.0, or **pI = 5.8**.



pH < 2.5 (charge = +2) | 2.5 < pH < 8.0 (charge = +1) | 8.0 < pH < 9.5 (charge = 0) | pH > 9.5 (charge = -1)

pI = 8.8, the average of 8.0 and 9.5.

2. My guesses, based on pI alone:

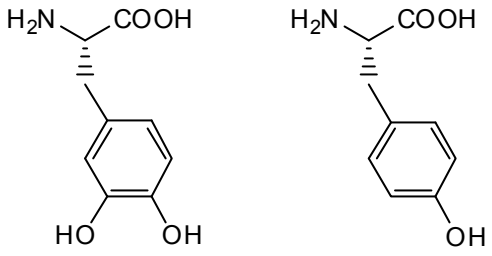


(eluted under gradient of increasing pH)

This doesn't take into account the effect of hydrophobicity/hydrophilicity.

3. Selenocysteine looks an awful lot like serine and cysteine, with the O/S replaced with a selenium atom. It is made by converting the -OH of serine into a good leaving group, followed by substitution.

L-DOPA is oxidized tyrosine:



L-DOPA tyrosine

Pyrrolysine is a combination of lysine and a methylated proline residue.