Calculating approximate isoelectric points for amino acids and peptides:

Amino acids:

If the side chain does not have an ionizable group, then the pI is simply the average of the α-NH_3 and α-COOH pKa values.

If the side chain has an ionizable group then all three pKa values must be considered.

- If the side chain is acidic (asp and glu), then average the sidechain pKa with the α-COOH pKa.
- If the side chain is basic (his, arg, and lys), then average the sidechain pKa with the α-NH_3 pKa.
- For other ionizable groups (tyr and cys), determine which is the middle pKa and average it with the α-COOH pKa.

Peptides:

A general rule of thumb that works for peptides (also works for amino acids):

Find the approximate pH where the net charge would be zero, and average the closest pKa values on either side of this value.

Examples:

Amino Acid	α-COOH	α-NH_3	pKa 3	Calc. pI
Gly	**2.35**	**9.78**	------	6.07
Asp	**2.10**	9.82	**3.86**	2.98
Cys	**1.86**	10.25	**8.00**	5.02
Tyr	**2.20**	**9.11**	10.07	5.66
Lys	2.18	**8.95**	**10.53**	9.74

Peptide	α-COOH	α-NH_3	pKa 3	pKa 4	Net Charge	Calc. pI
Gly-Asp-Glu	2.10 (glu)	9.78 (gly)	**3.86 (asp)**	**4.07 (glu)**		3.97
Charge at pH 1	0	+1	0	0	+1	
2	0	+1	0	0	+1	
3	-1	+1	0	0	0	
4	-1	+1	-1	0	-1	
5	-1	+1	-1	-1	-2	
6	-1	+1	-1	-1	-2	
7	-1	+1	-1	-1	-2	
8	-1	+1	-1	-1	-2	
9	-1	+1	-1	-1	-2	
10	-1	0	-1	-1	-3	

Peptide	α-COOH	α-NH_3	pKa 3	pKa 4	pKa 5	pKa 6	Net Charge	Calc. pI
Cys-Glu-Lys-Arg	2.01 (arg)	**10.25 (cys)**	**8.00 (cys)**	4.07 (glu)	10.53 (lys)	12.48 (arg)		9.13
Charge at pH 1	0	+1	0	0	+1	+1	+3	
2	0	+1	0	0	+1	+1	+3	
3	-1	+1	0	0	+1	+1	+2	
4	-1	+1	0	0	+1	+1	+2	
5	-1	+1	0	-1	+1	+1	+1	
6	-1	+1	0	-1	+1	+1	+1	
7	-1	+1	0	-1	+1	+1	+1	
8	-1	+1	0	-1	+1	+1	+1	
9	-1	+1	-1	-1	+1	+1	-1	
10	-1	+1	-1	-1	+1	+1	-1	
11	-1	0	-1	-1	0	+1	-2	
12	-1	0	-1	-1	0	+1	-2	
13	-1	0	-1	-1	0	0	-3	