IIT Guwahati

Lecture 4

Course BT 631

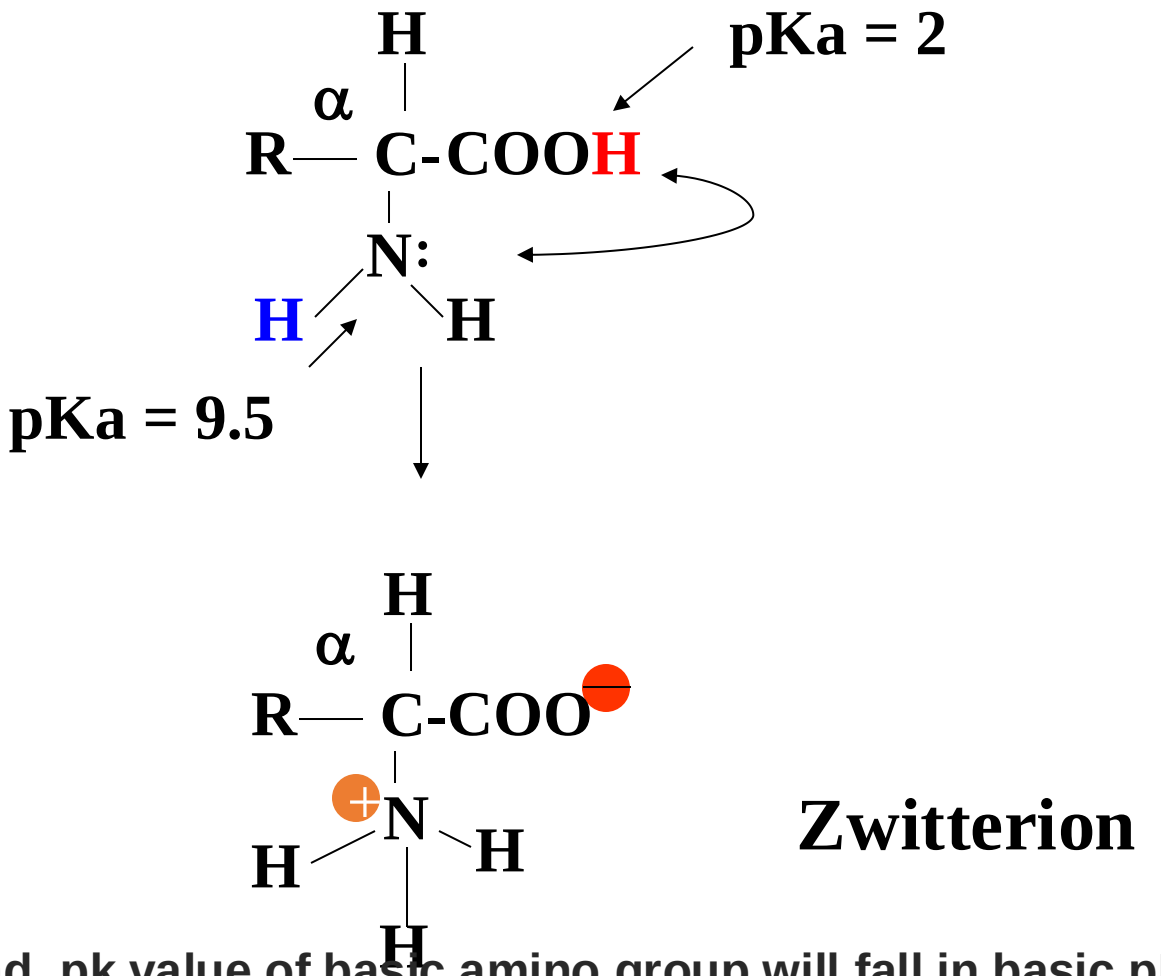
Protein Structure function and Crystallography

Prof. Arun Goyal

Dept. of Biosciences and Bioengineering

Ionization state of amino Acid depends on pH

Charges on Amino Acids

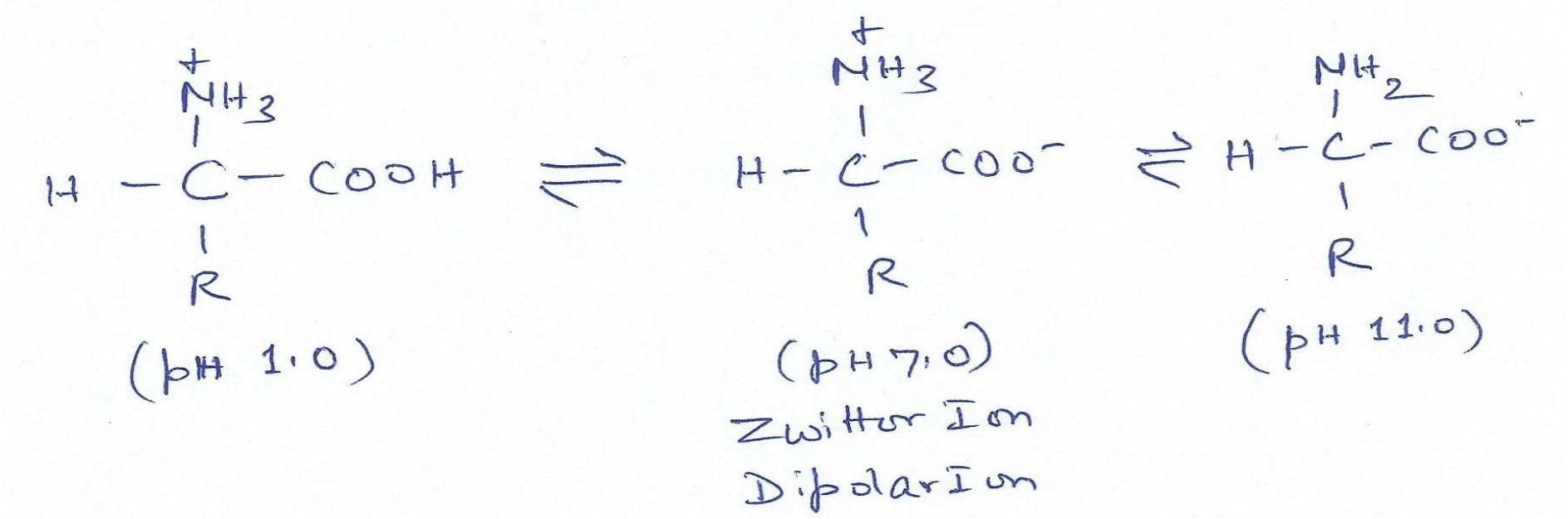


Zwitterion

pk value of acid group will fall in acidic pH and, pk value of basic amino group will fall in basic pH range.

Ionization state of amino Acid depends on pH

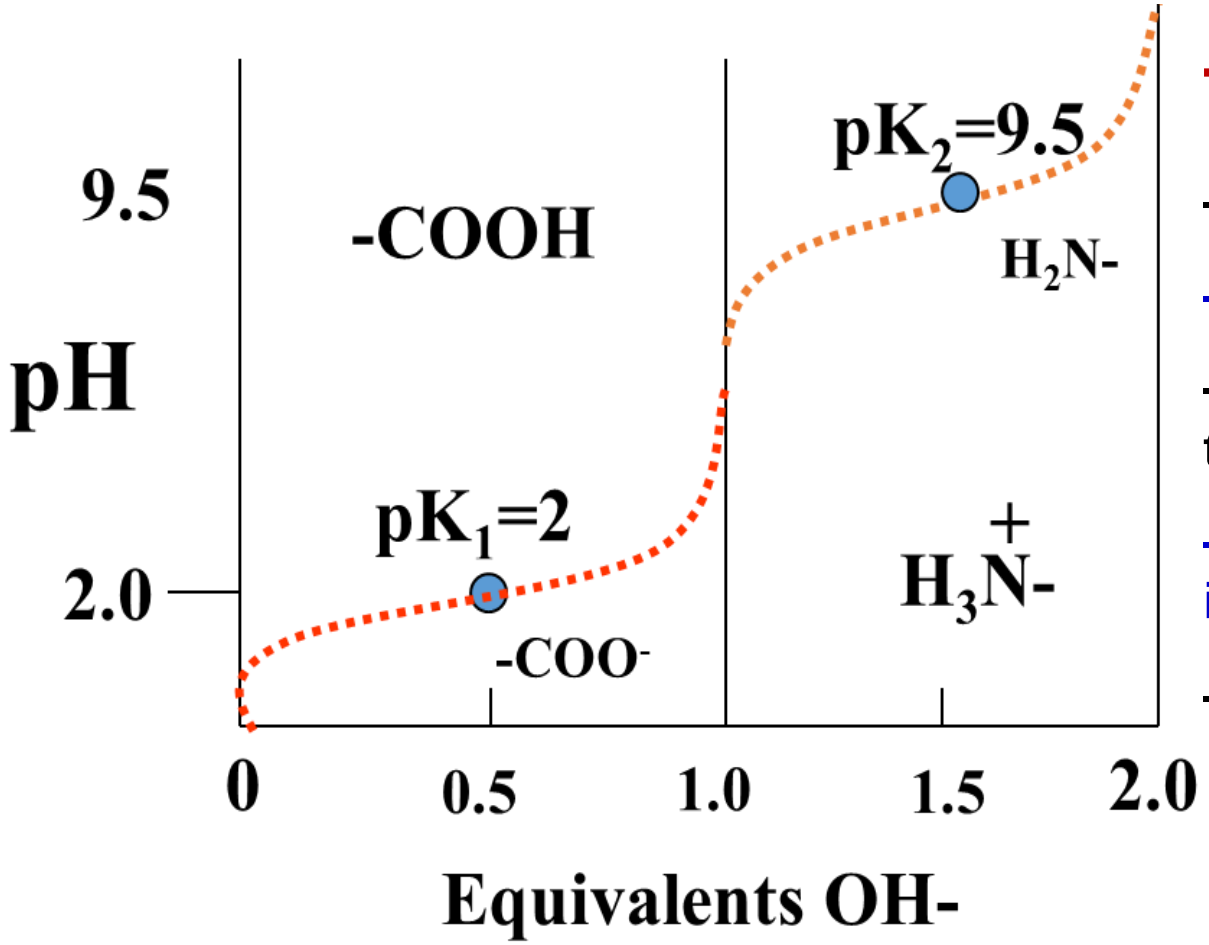
A general Amino acid exists as Zwittor ion (Diploar ion) at Neutral pH



A zwitterion also called an inner salt, is a molecule that contains an equal number of positively- and negatively-charged functional groups and the net charge of the entire molecule is zero.

Titration curve of an Amino acid

To draw a titration curve, 1) Determine number of dissociable groups, 2) place pK values in center of range (3) draw curve.



Titration curve for glycine

– Note that one starts with all groups in acid form.

– Note how many equivalents are added?

– Note that at 0.5 and 1.5 equivalents, pH is equal to pK of group being titrated.

– Note pH which gives zero charge is the isoelectric point. Calculated as (pK1+ pK2)/2

– Note where the buffering capacity is best?

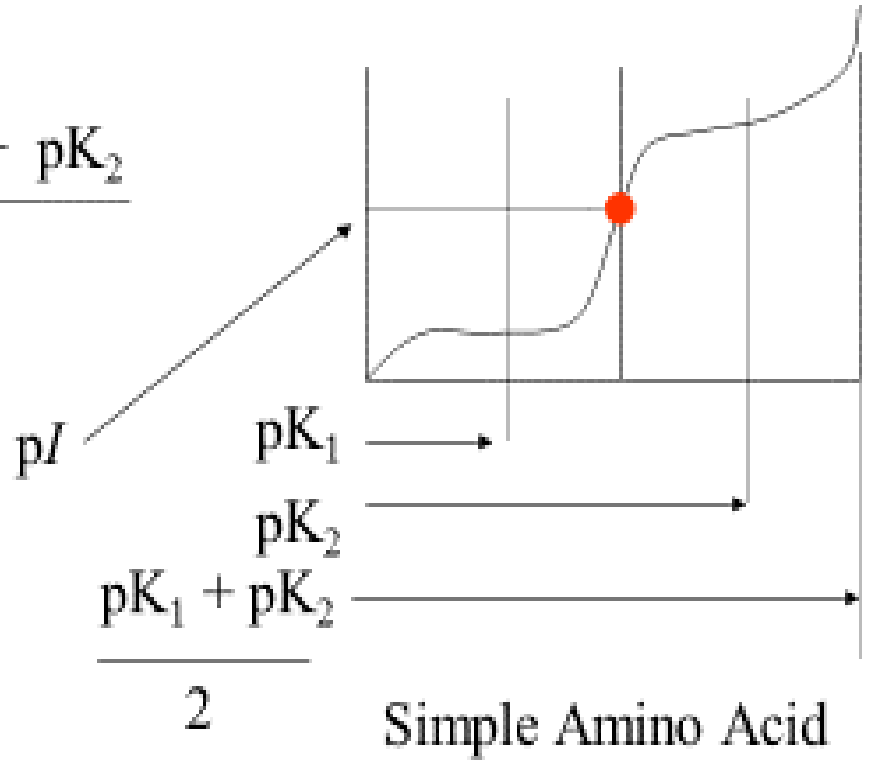
Isoelectric Point (or Isoionic Point) of Amino Acids

Isoelectric Point (pI)

It is the pH at which the amino acid does not migrate i.e. it is the pH at which amino acid is neutral i.e. in Zwitter ion or Dipolar form.

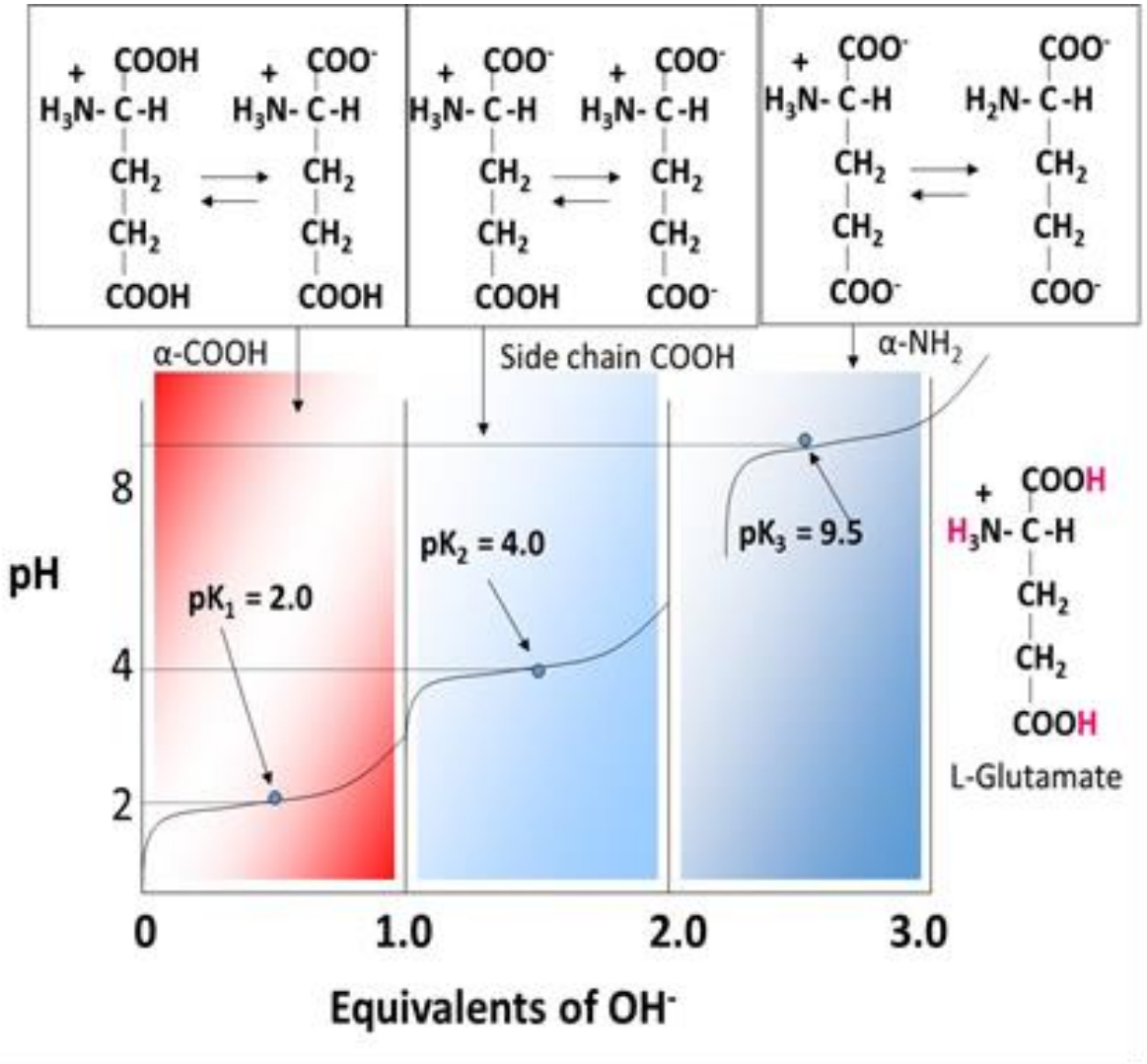
1. pH at neutral charge

2. $pI = \frac{pK_1 + pK_2}{2}$



pI of Glycine = (9.5+2.0)/2=11.5/2 = 5.75

Isoelectric point of Glutamate (Acidic amino acid)



The pI of glutamate will be at lower pH because the acidic side chain introduces and extra negative charge. *So the neutral form exists under more acidic conditions* when the extra negative charge has been neutralized.

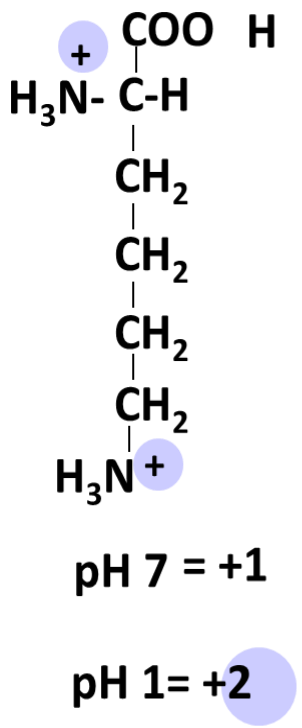
$$pI = \frac{pK_1 + pK_2}{2} = \frac{2.0 + 4.0}{2} = 3.0$$

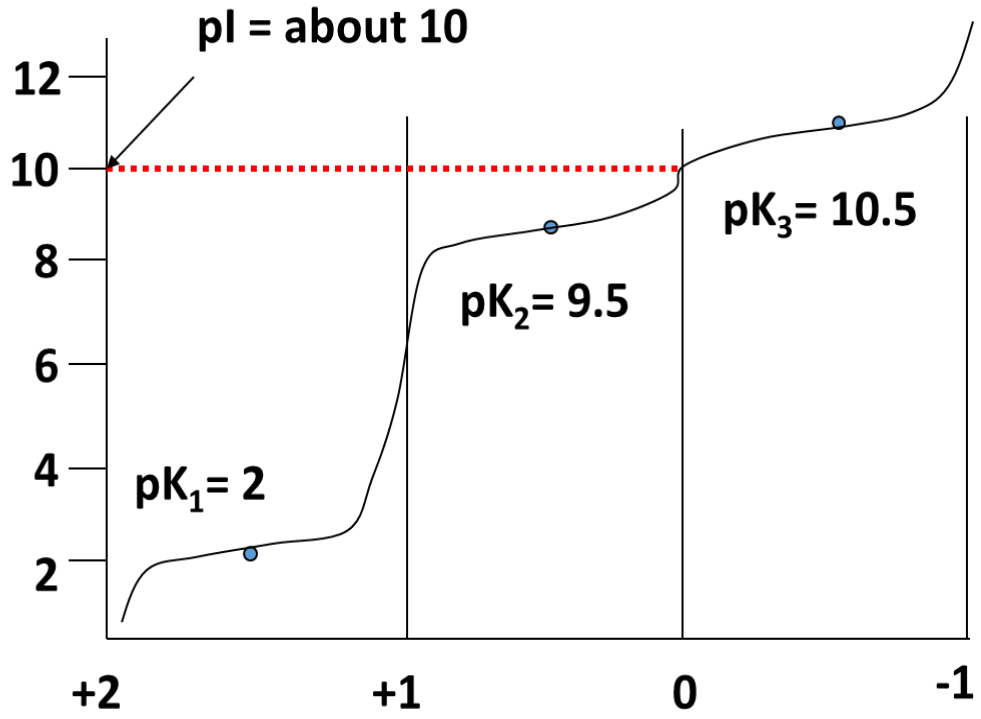
Rule: Always use the 2 pKs that border the "0" charged species.

Isoelectric point of Lysine (Basic Amino acid)

The pI of Lysine will be at higher pH because the basic side chain introduces an extra positive charge.

So the neutral form exists under more basic conditions **when the extra positive charge has been neutralized (by more OH^- ions).**





Lysine is a basic amino acid

$$pI = \frac{pK_2 + pK_3}{2} = \frac{9.5 + 10.5}{2} = 10$$

pK values of Ionizable groups in Amino acids

Amino Acid	Abbreviations		pK_1	pK_2	pK_3
Glycine	Gly	G	2.34	9.6	
Alanine	Ala	A	2.34	9.69	
Valine	Val	V	2.32	9.62	
Leucine	Leu	L	2.36	9.6	
Isoleucine	Ile	I	2.36	9.6	
Serine	Ser	S	2.21	9.15	
Threonine	Thr	T	2.63	10.43	
Methionine	Met	M	2.28	9.21	
Phenylalanine	Phe	F	1.83	9.13	
Tryptophan	Trp	W	2.83	9.39	
Asparagine	Asn	N	2.02	8.8	
Glutamine	Gln	Q	2.17	9.13	
Proline	Pro	P	1.99	10.6	
Cysteine	Cys	C	1.71	10.78	8.33
Histidine	His	H	1.82	9.17	6.0
Aspartic acid	Asp	D	2.09	9.82	3.86
Glutamic acid	Glu	E	2.19	9.67	4.25
Tyrosine	Tyr	Y	2.2	9.11	10.07
Lysine	Lys	L	2.18	8.95	10.79
Arginine	Arg	R	2.17	9.04	12.48
			2.20	9.52	