

Metabolic Classification of the Amino Acids

***Essential and Non-essential**

*** Glucogenic and Ketogenic**

Essential Amino Acids

- Of the 20 amino acids that make up proteins 10 of them can be synthesized by the human body
- The other 10 amino acids **must be acquired from food** sources. These amino acids are known as **essential amino acids**

Essential Amino Acids

Complete protein

- **Contains all 10 essential amino acids**
- **Proteins derived from animal sources are complete proteins**
- **Beans contain some complete protein as well**

Incomplete protein

- **Lack one or more of the essential amino acids**
- **Most vegetable proteins are incomplete proteins**
- **Beans are an exception to this generalization**

Essential Amino Acids in Humans

- **Required in diet**
- **Humans incapable of forming requisite carbon skeleton**

- **Arginine***
- **Histidine***
- **Isoleucine**
- **Leucine**
- **Valine**
- **Lysine**
- **Methionine**
- **Threonine**
- **Phenylalanine**
- **Tryptophan**

*** Essential in children, not in adults**

Non-Essential Amino Acids in Humans

- Not required in diet
- Can be formed from α -keto acids by transamination and subsequent reactions
 - Alanine
 - Asparagine
 - Aspartate
 - Glutamate
 - Glutamine
 - Glycine
 - Proline
 - Serine
 - Cysteine (from Met^{*})
 - Tyrosine (from Phe^{*})

* Essential amino acids

Essential and Nonessential Amino Acids

Nonessential	Essential
Alanine	Arginine*
Asparagine	Histidine*
Aspartate	Isoleucine
Cysteine	Leucine
Glutamate	Lysine
Glutamine	Methionine
Glycine	Phenylalanine
Proline	Threonine
Serine	Tryptophan
Tyrosine	Valine

Amino acids are classified as glucogenic or ketogenic

- ❑ **Glucogenic amino acids** are degraded to compounds that can be used as carbon skeletons for glucose synthesis via gluconeogenesis

Ketogenic amino acids are degraded to compounds that can only be used to generate the ketone bodies

Both Glucogenic and ketogenic amino acids:
Several amino acids are classified as both glucogenic and ketogenic because of their degradation products

Glucogenic Amino Acids

- Metabolized to α -ketoglutarate, pyruvate, oxaloacetate, fumarate, or succinyl CoA
  Phosphoenolpyruvate  Glucose

- Aspartate
- Asparagine
- Arginine
- Phenylalanine
- Tyrosine
- Isoleucine

- Methionine
- Valine
- Glutamine
- Glutamate
- Proline
- Histidine

- Alanine
- Serine
- Cysteine
- Glycine
- Threonine
- Tryptophan

Ketogenic Amino Acids

- Metabolized to acetyl CoA or acetoacetyl CoA

Animals cannot convert acetyl CoA or acetoacetyl CoA to pyruvate

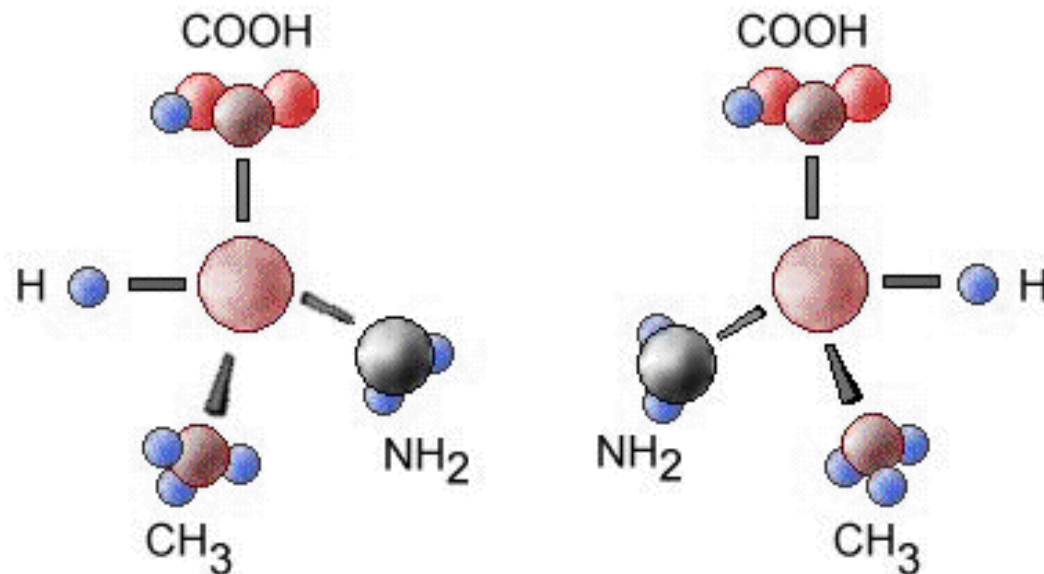
- **Leucine** *
- **Lysine** *
- Tryptophan
- Phenylalanine
- Tyrosine
- Isoleucine
- Threonine

* Leucine and lysine are only ketogenic

Properties of Amino Acids

Amino Acids and Optical Isomers

- Except for glycine, all amino acids have a **chiral carbon atom**. Therefore they can have **optical isomers**
- The amino acids found in **proteins** are all **levorotatory or L forms**.



Optical properties of amino acids

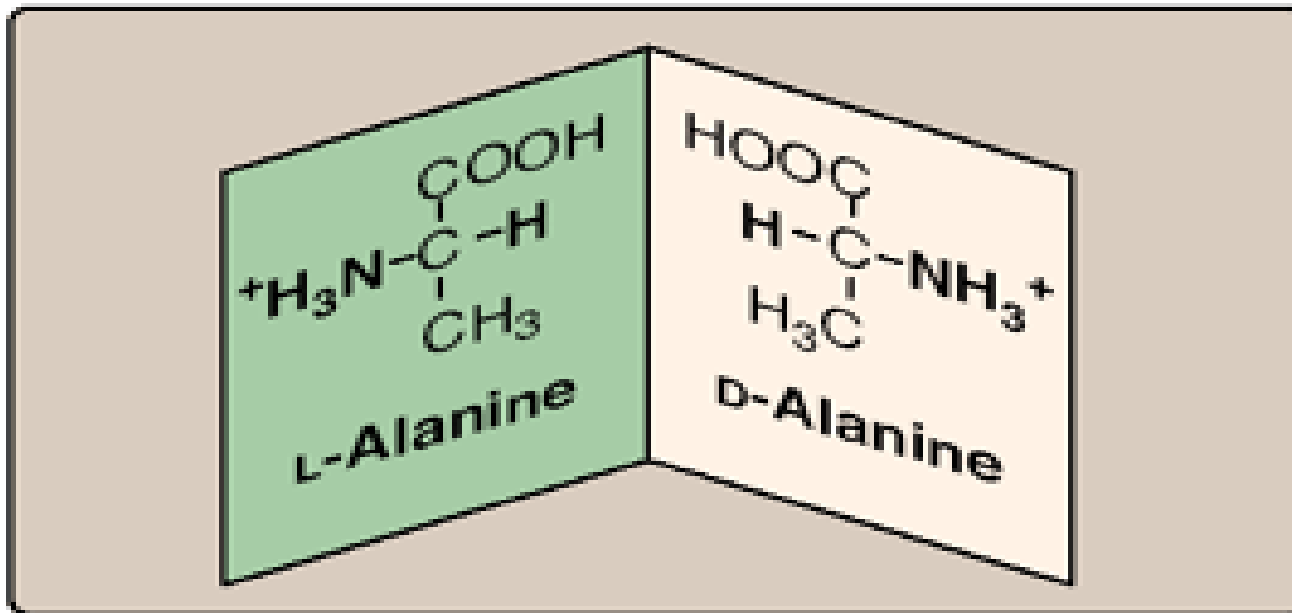
The α -carbon of each amino acid is attached to four different chemical groups and is, therefore, a chiral or optically active carbon atom.

Glycine is the exception because its α -carbon has two hydrogen substituents and, therefore, is optically inactive.

Amino acids that have an asymmetric center at the α -carbon can exist in two forms, designated D and L, that are mirror images of each other. The two forms in each pair are termed **stereoisomers**, optical isomers, or **enantiomers**.

All amino acids found in proteins are of the **L-configuration**.

However, D-amino acids are found in some antibiotics and in plant and bacterial cell walls.



D and L forms of alanine are mirror images.

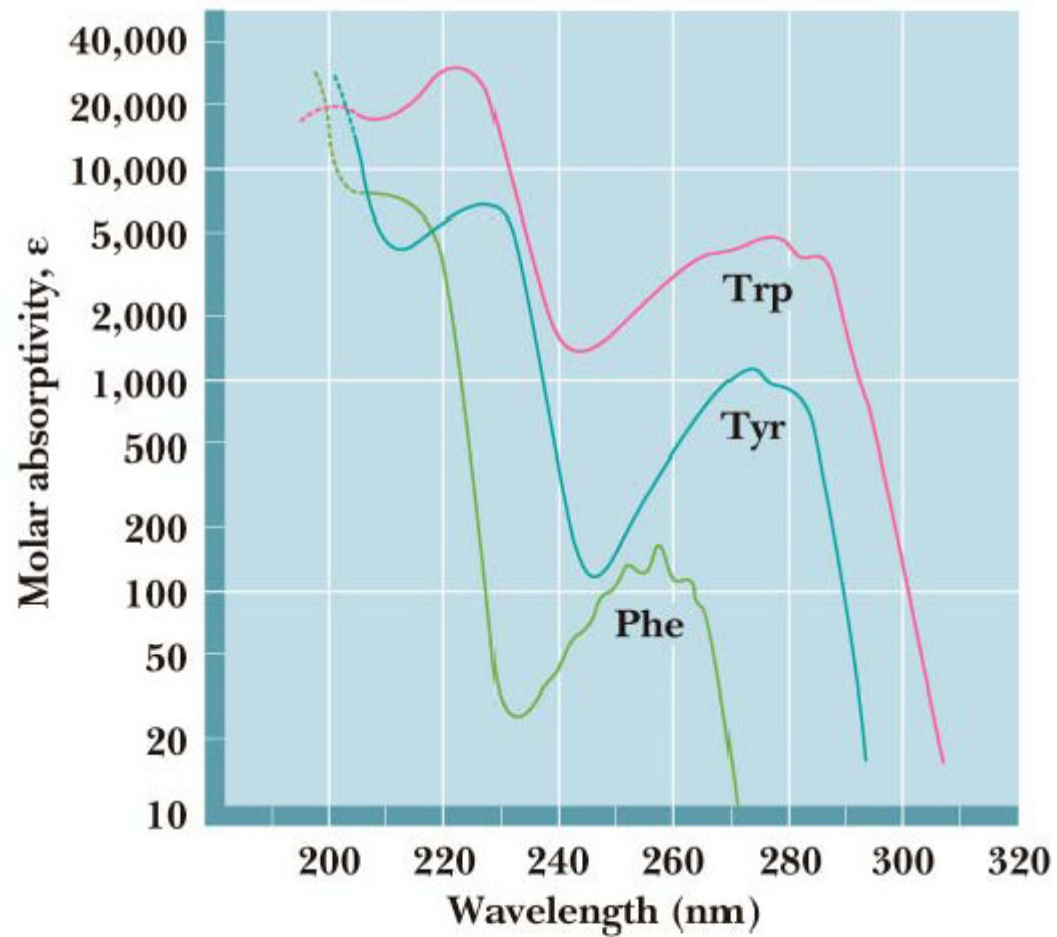
-So :

- 19 of the 20 common amino acids have a chiral α -carbon atom (Gly does not)
- **Amino acids have stereoisomers (except glycine) .**
- Threonine and isoleucine have 2 chiral carbons each (4 possible stereoisomers each)
- Mirror image pairs of amino acids are designated L (levo) and D (dextro)
- Proteins are assembled from L-amino acids (a few D-amino acids occur in nature)

Spectroscopy of Amino Acids

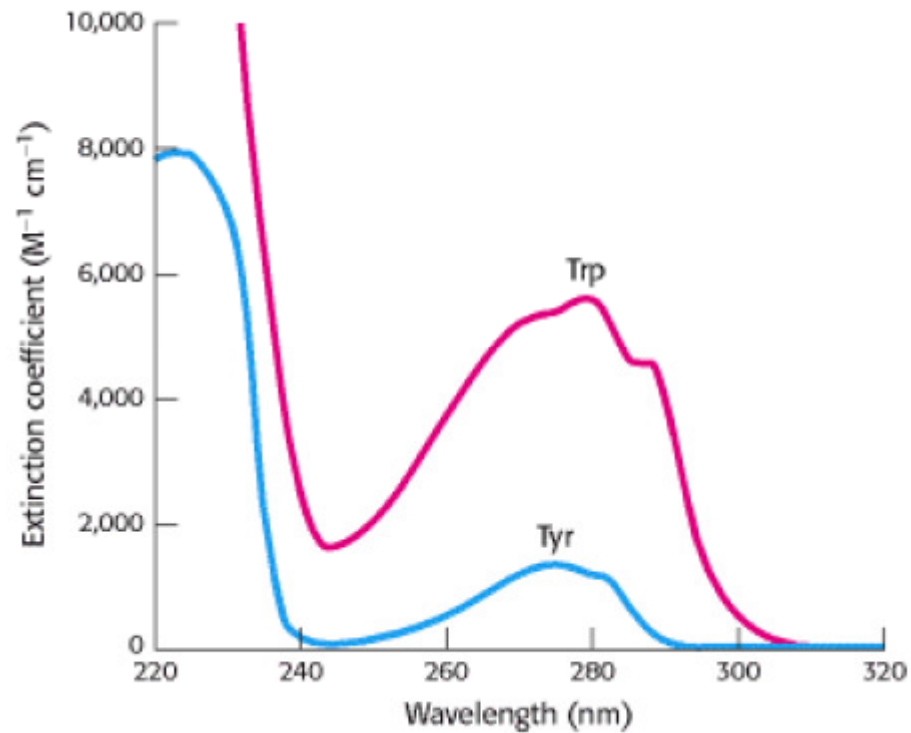
- The aromatic R-groups in amino acids absorb ultraviolet light with an absorbance maximum in the range of **280nm**.
- Only phenylalanine, tyrosine and tryptophan (**aromatic amino acids**) can absorb UV light . The ability of proteins to absorb ultraviolet light is predominantly due to the presence of the tryptophan which strongly absorbs ultraviolet light.

Spectroscopy of amino acids



Only the aromatic amino acids absorb light in the UV region

Only these



Absorption Spectra of the Aromatic Amino Acids Tryptophan (Red) and Tyrosine (Blue).

*For tryptophan, absorption is maximum at 280 nm whereas, for tyrosine, absorption is maximum at 276 nm .

*Phenylalanine absorbs light less strongly and at shorter wavelengths.

*The absorption of light at 280 nm can be used to estimate the concentration of a protein in solution if the number of tryptophan and tyrosine residues in the protein is known.

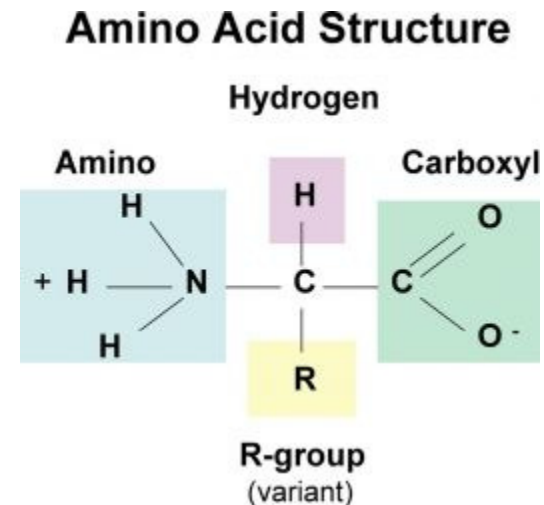
Acidic and Basic Properties of Amino Acids

*Amino acids in aqueous solution contain weakly acidic α -carboxyl groups and weakly basic α -amino groups. In addition, each of the acidic and basic amino acids contains an ionizable group in its side chain.

*Thus, both free amino acids and some amino acids combined in peptide linkages can act as buffers.

*Recall that acids may be defined as proton donors and bases as proton acceptors. Acids (or bases) described as “weak” ionize to only a limited extent.

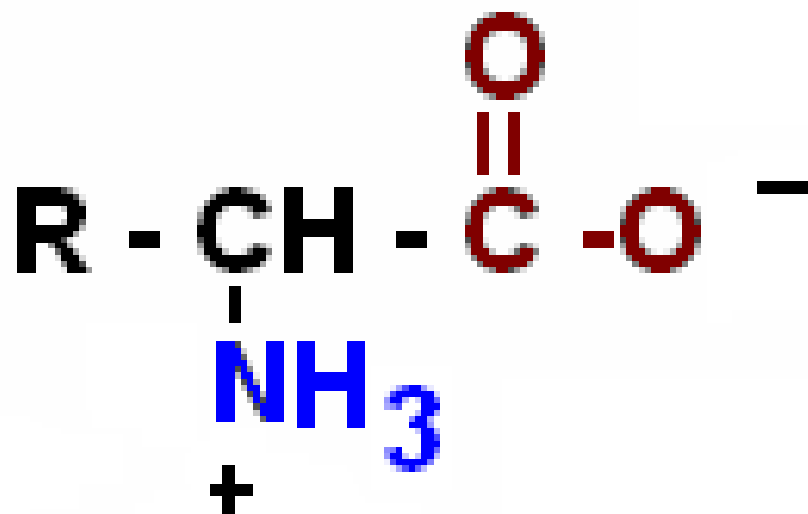
- At physiological pH (around 7.4) the carboxyl group will be unprotonated and the amino group will be protonated.



- An amino acid with **no ionizable R-group** would be **electrically neutral** at this pH.
- This species is termed a **zwitterion**.
- It is polar but has no net charge.

The Zwitterion

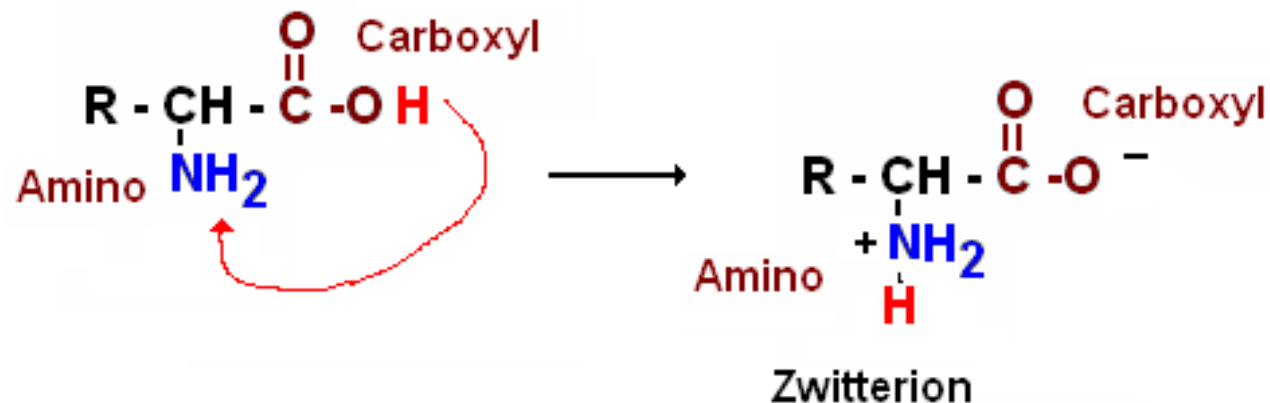
This dipolar ion form is known as a **Zwitterion**



Zwitterion

Amino Acids are Amphoteric

- So , Amino acids are **amphoteric**. They are capable of behaving as both an acid and a base, since they have both a proton donor group and a proton acceptor group.



- The α -COOH and α -NH₂ groups in amino acids are capable of ionizing (as are the acidic and basic R-groups of the amino acids).
- As a result of their ionizability the following ionic equilibrium reactions may be written:
- **$\text{R-COOH} \rightleftharpoons \text{R-COO}^- + \text{H}^+$**
- **$\text{R-NH}_3^+ \rightleftharpoons \text{R-NH}_2 + \text{H}^+$**

The acidic strength of the carboxyl, amino and ionizable R-groups in amino acids can be defined by the dissociation constant, K_a or more commonly the negative logarithm of K_a , the pK_a .

The **net charge** (the algebraic sum of all the charged groups present) of any amino acid, peptide or protein, will depend upon the pH of the surrounding aqueous environment.

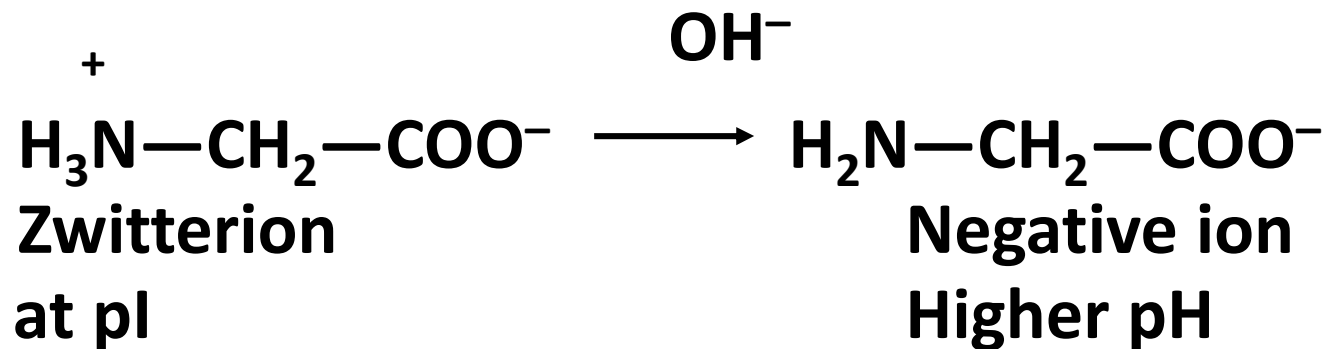
- As the pH of a solution of an amino acid changes , the net charge of amino acid will change .
- This phenomenon can be observed during the titration of any amino acid or protein.
- When the net charge of an amino acid or protein is zero the pH will be equivalent to the **isoelectric point: pI**.

Isoelectric Point (pI)

Isoelectric pH is that **pH** when the negative charge is equal to the positive charge.

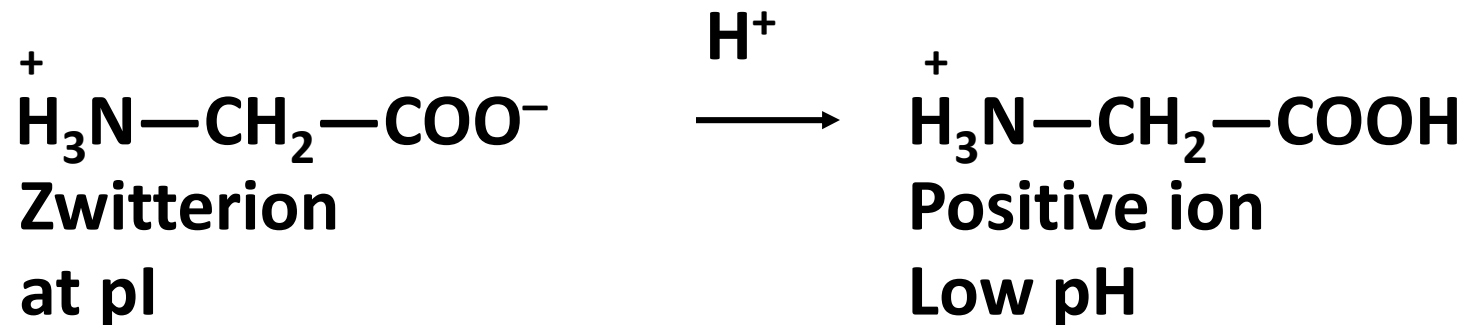
Amino Acids as Acids

- In solutions more basic than the pI, the —NH_3^+ in the amino acid donates a proton.



Amino Acids as Bases

- In solution more acidic than the pI, the COO⁻ in the amino acid accepts a proton.

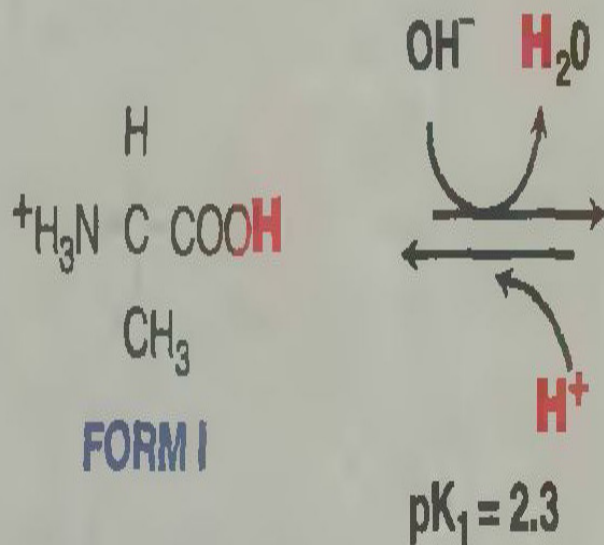


Titration of an amino acid

Dissociation of the carboxyl group: The titration curve of an amino acid can be analyzed in the same way as weak acid. Consider alanine, for example, which contains both an α -carboxyl and an α -amino group.

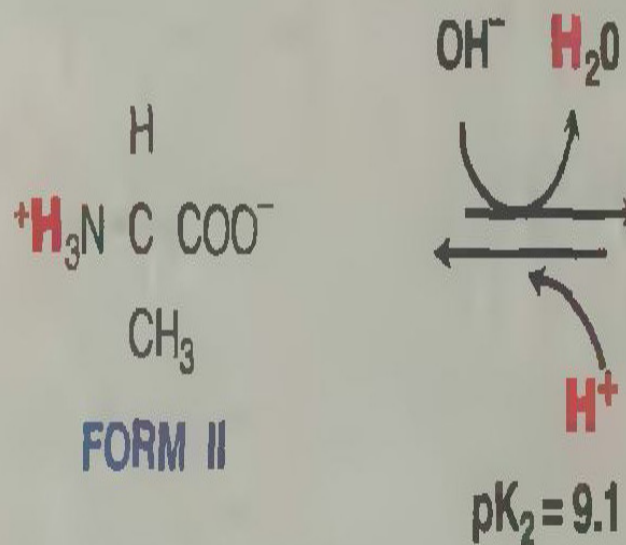
At a low (acidic) pH, both of these groups are protonated (shown in Figure).

As the pH of the solution is raised, the -COOH group of Form I can dissociate by donating a proton to the medium. The release of a proton results in the formation of the carboxylate group, -COO^- . This structure is shown as Form II, which is the dipolar form of the molecule . This form, also called a **zwitterion**, is **the isoelectric form** of alanine—that is, it has an overall charge of zero.



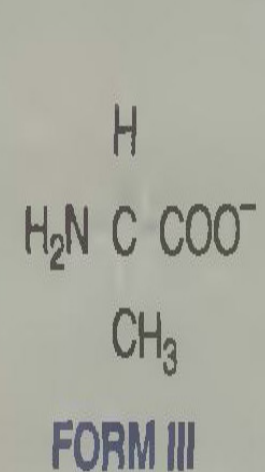
Alanine in acid solution
(pH less than 2)

Net charge = +1



Alanine in neutral solution
(pH approximately 6)

Net charge = 0
(isoelectric form)



Alanine in basic solution
(pH greater than 10)

Net charge = -1

Application of the Henderson-Hasselbalch equation:

The dissociation constant of the carboxyl group of an amino acid is called K_1 , because the molecule contains a second titratable group.

The Henderson-Hasselbalch equation can be used to analyze the dissociation of the carboxyl group of alanine in the same way as described for weak acid:

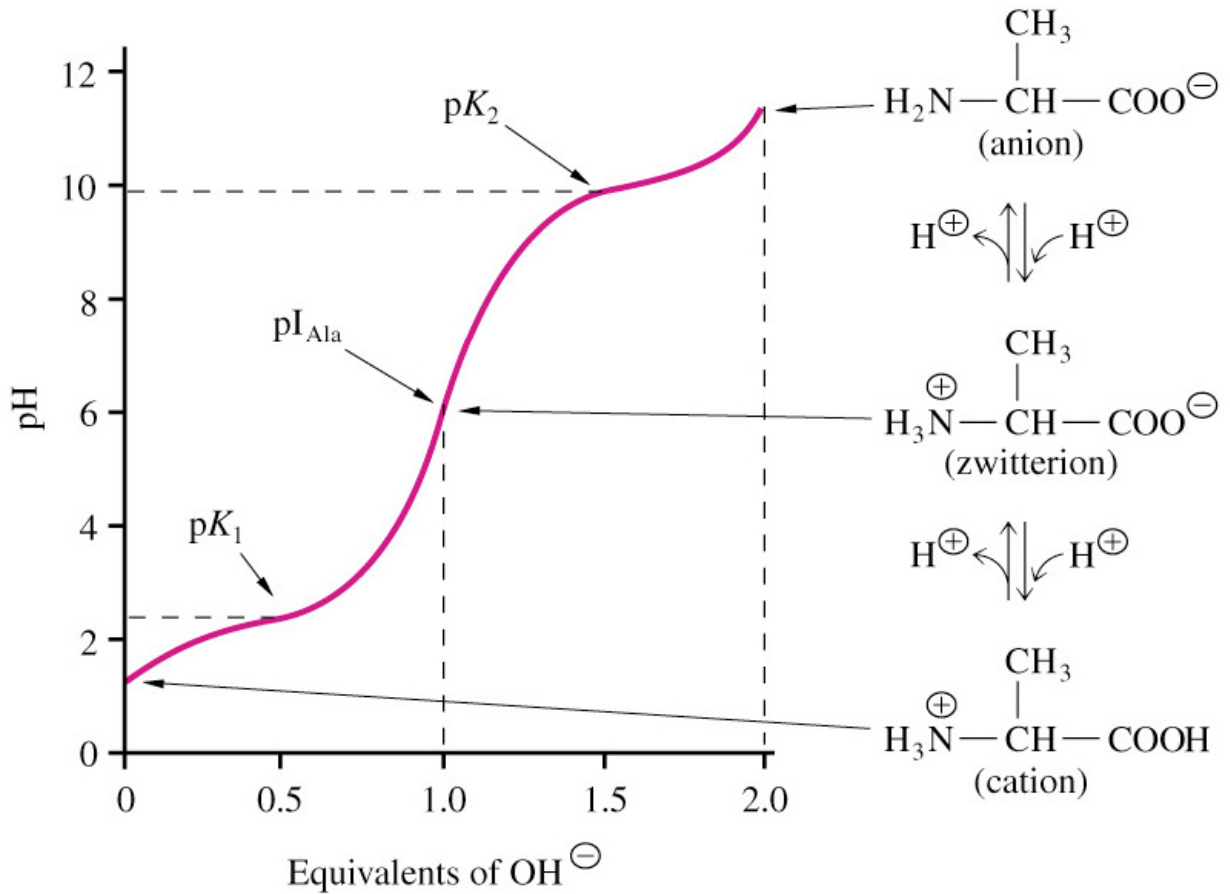
$$K_1 = \frac{[H^+][II]}{[I]}$$

where I is the fully protonated form of alanine, and II is the isoelectric form of alanine. This equation can be rearranged and converted to its logarithmic form to yield:

$$pH = pK_1 + \log \frac{[II]}{[I]}$$

Titration curve for alanine

- Titration curves are used to determine pK_a values
- $pK_1 = 2.4$
- $pK_2 = 9.9$
- $pI_{Ala} =$ isoelectric point
- $pI = (pK_1 + pK_2) / 2 = 6.1$



Dissociation of the amino group:

*The second titratable group of alanine is the amino ($-\text{NH}_3^+$) group . This is a much weaker acid than the $-\text{COOH}$ group and, therefore, has a much smaller dissociation constant, K_2 . [Note: Its pK_a is therefore larger.]

*Release of a proton from the protonated amino group of Form II results in the fully deprotonated form of alanine, Form III .

*Each titratable group has a pK_a that is numerically equal to the pH at which exactly one half of the protons have been removed from that group.

***The pK_a for the most acidic group ($-\text{COOH}$) is pK_1 , whereas the pK_a for the next most acidic group ($-\text{NH}_3^+$) is pK_2 .**

Titration curve of alanine

By applying the Henderson-Hasselbalch equation to each dissociable acidic group, it is possible to calculate the complete titration curve of a weak acid.

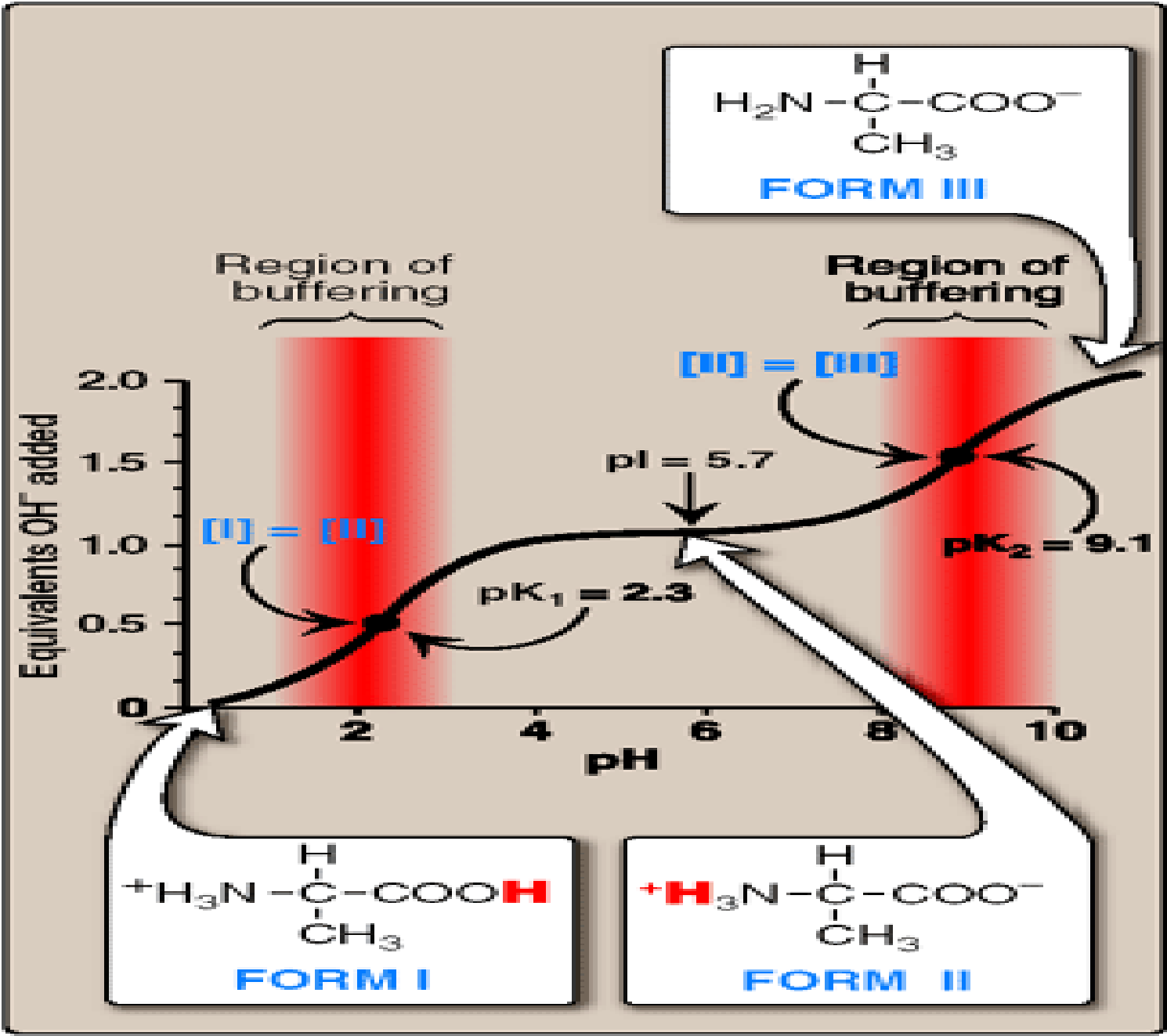
The Figure shows the change in pH that occurs during the addition of base to the fully protonated form of alanine (I) to produce the completely deprotonated form (III).

Note the following:

Buffer pairs: The **-COOH/-COO⁻** pair can serve as a buffer in the pH region around pK_1 , and the **-NH₃⁺/-NH₂** pair can buffer in the region around pK_2 .

When pH=pK: When the pH is equal to pK_1 (2.3), equal amounts of Forms I and II of alanine exist in solution.

When the pH is equal to pK_2 (9.1), equal amounts of Forms II and III are present in solution.



Isoelectric point: At neutral pH, alanine exists predominantly as the dipolar Form II in which the amino and carboxyl groups are ionized, but the **net charge is zero**.

The isoelectric point (pI) is **the pH at which an amino acid is electrically neutral— that is, in which the sum of the positive charges equals the sum of the negative charges**.

For an amino acid, such as alanine, that has only two dissociable hydrogens (one from the α -carboxyl and one from the α -amino group), **the pI is the average of pK_1 and pK_2 ($pI = [2.4 + 9.9]/2 = 6.15$)** The pI is thus midway between pK_1 (2.4) and pK_2 (9.9). pI corresponds to the pH at which the Form II (with a net charge of zero) predominates, and at which there are also equal amounts of Forms I (net charge of +1) and III (net charge of -1).

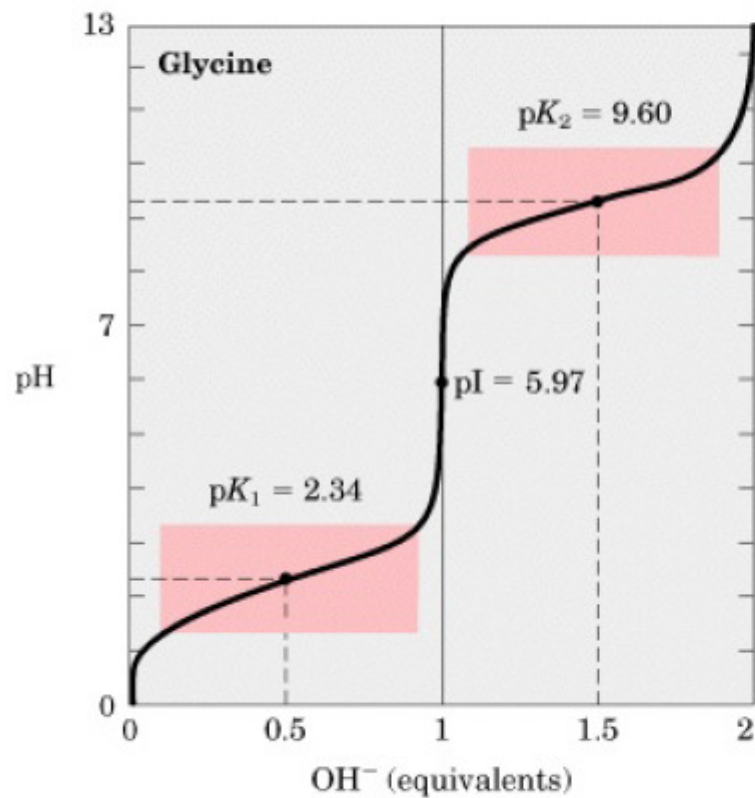
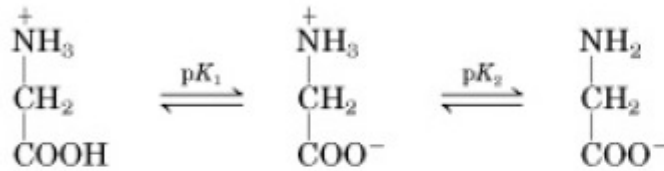
Net charge of amino acids at neutral pH:

*At physiologic pH, all amino acids have a negatively charged group ($-\text{COO}^-$) and a positively charged group ($-\text{NH}_3^+$), both attached to the α -carbon.

*Glutamate, aspartate, histidine, arginine, and lysine have additional potentially charged groups in their side chains.

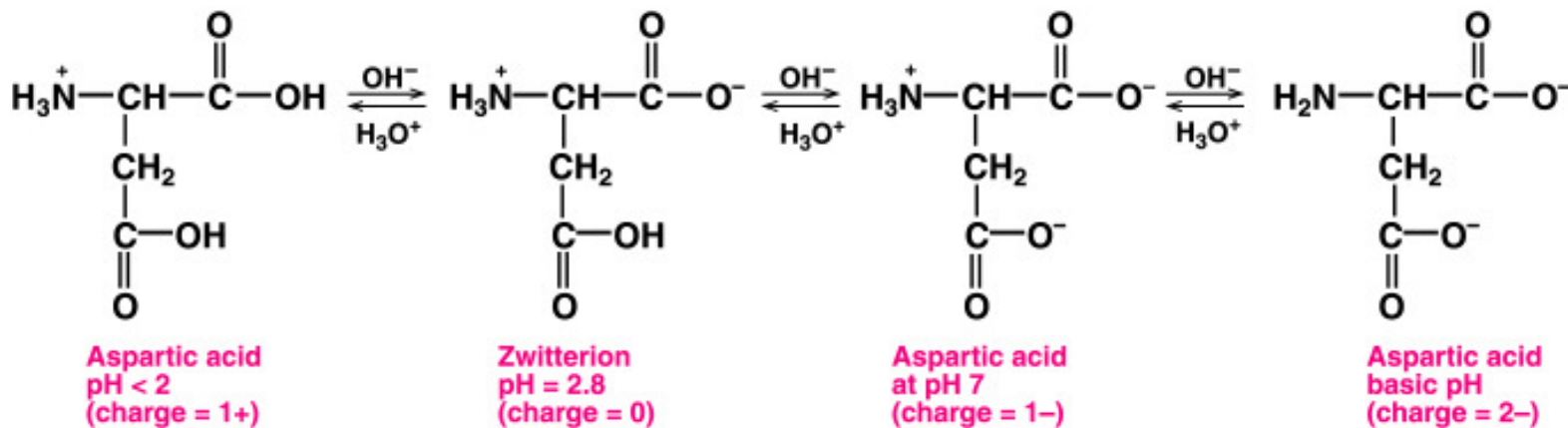
*Substances, such as amino acids, that can act either as an acid or a base are defined as **amphoterics**, and are referred to as ampholytes (amphoterics electrolytes).

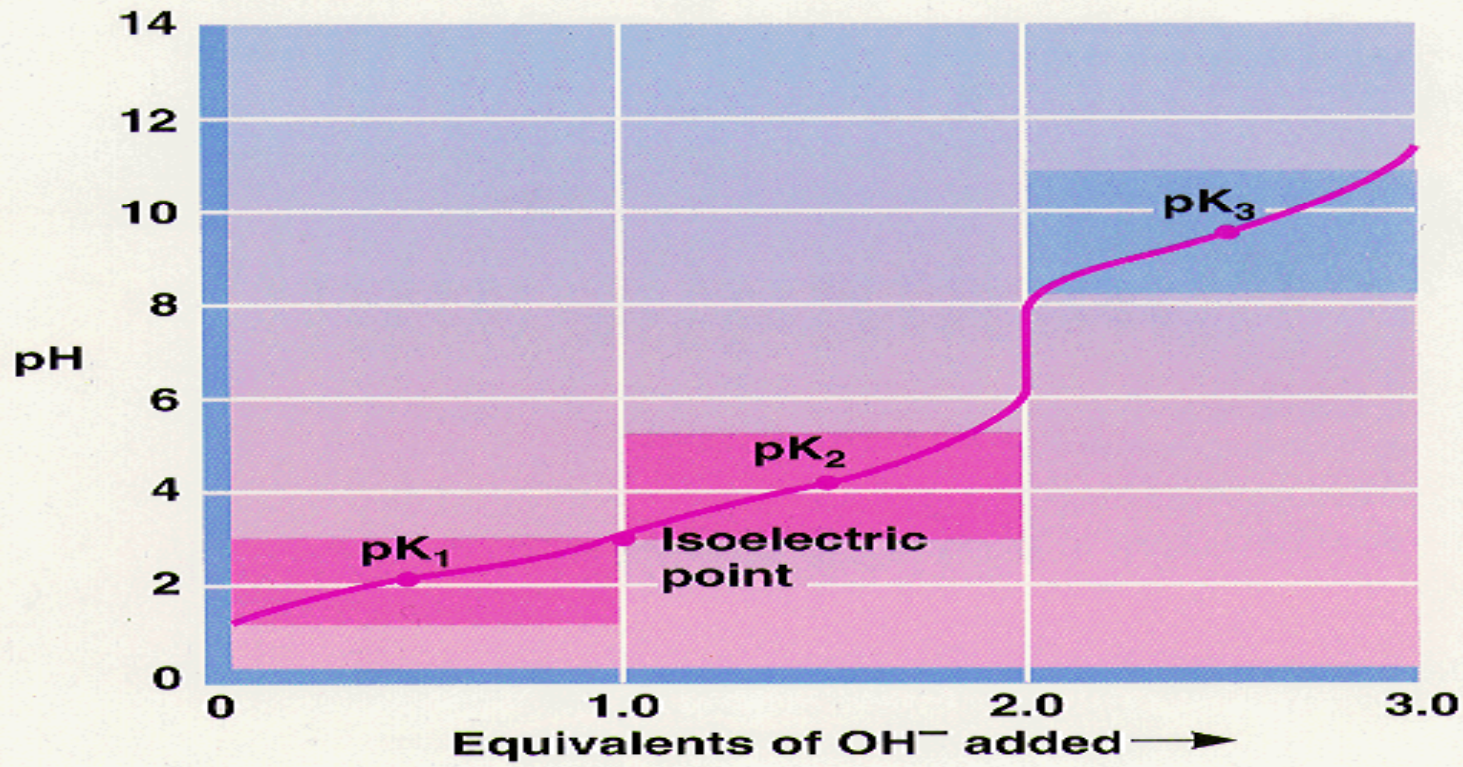
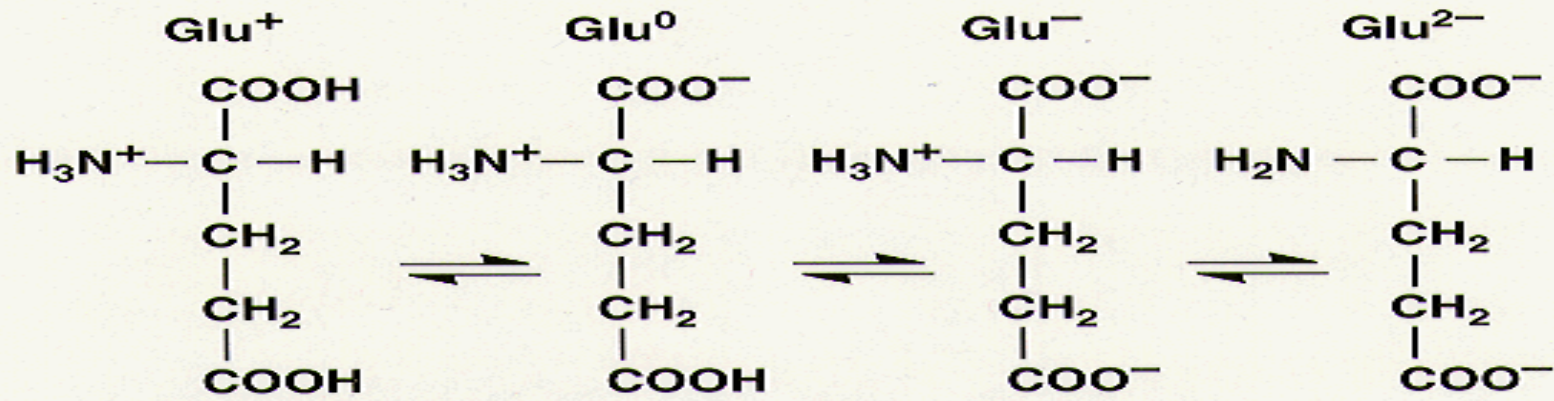
Acid-Base Properties of Amino Acids

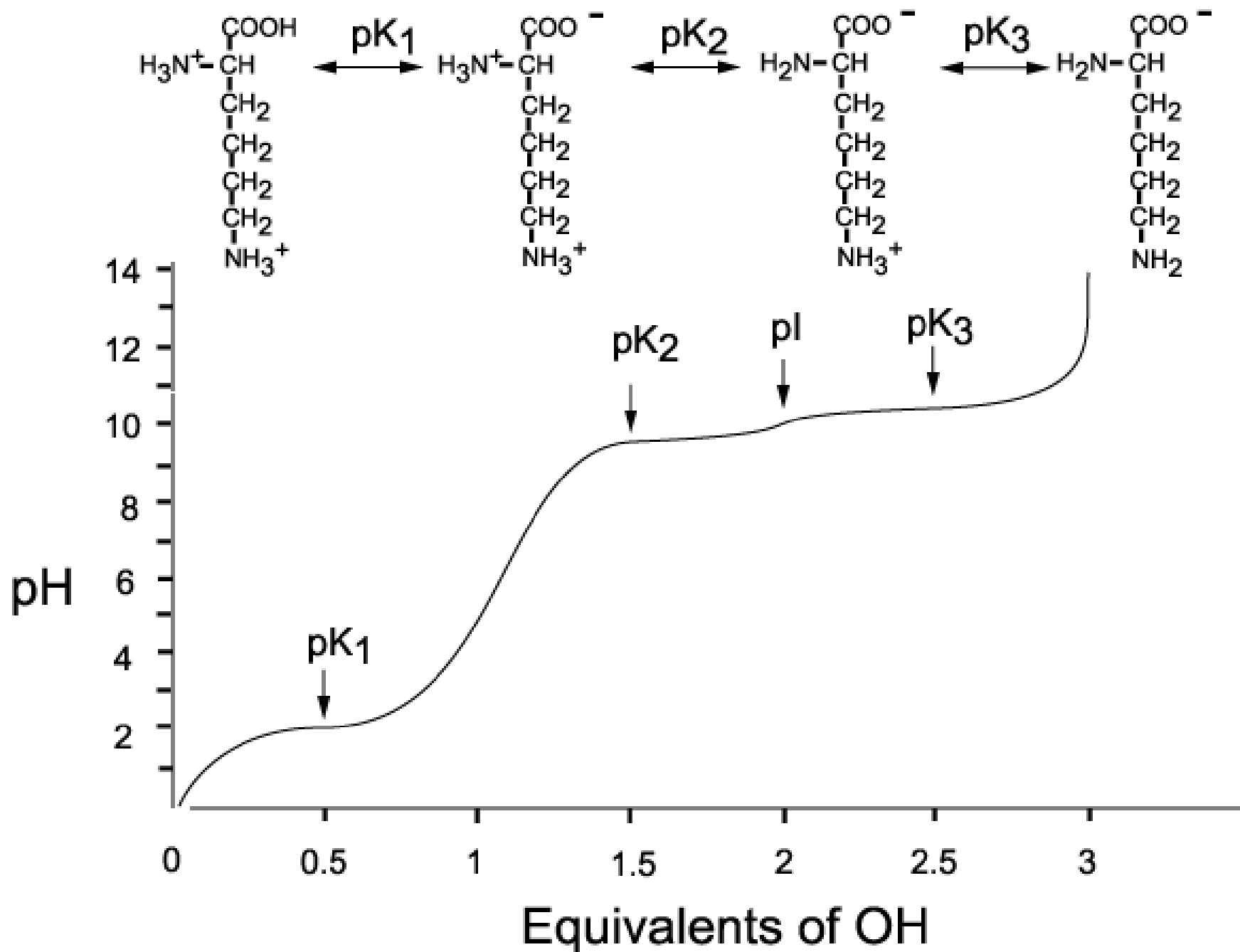


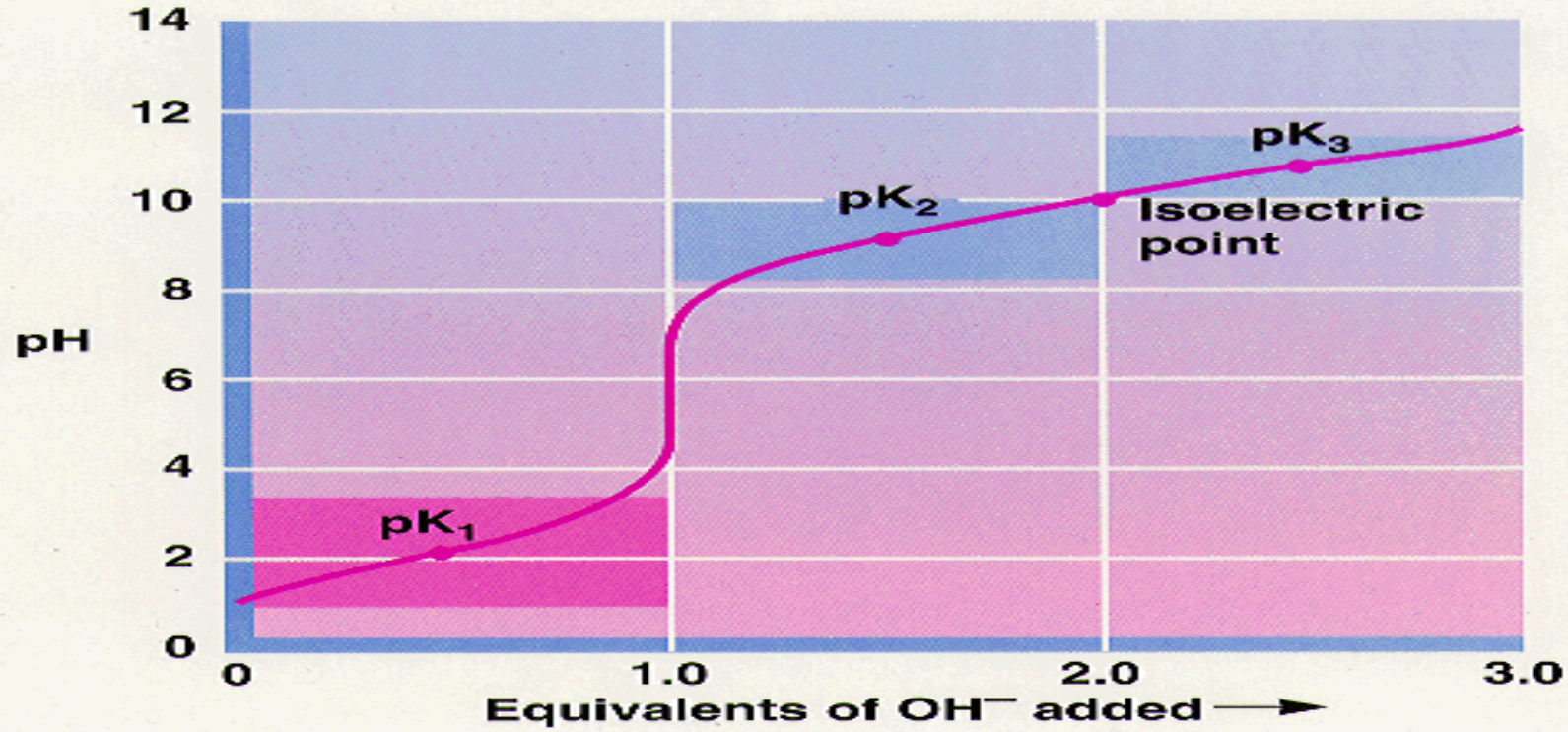
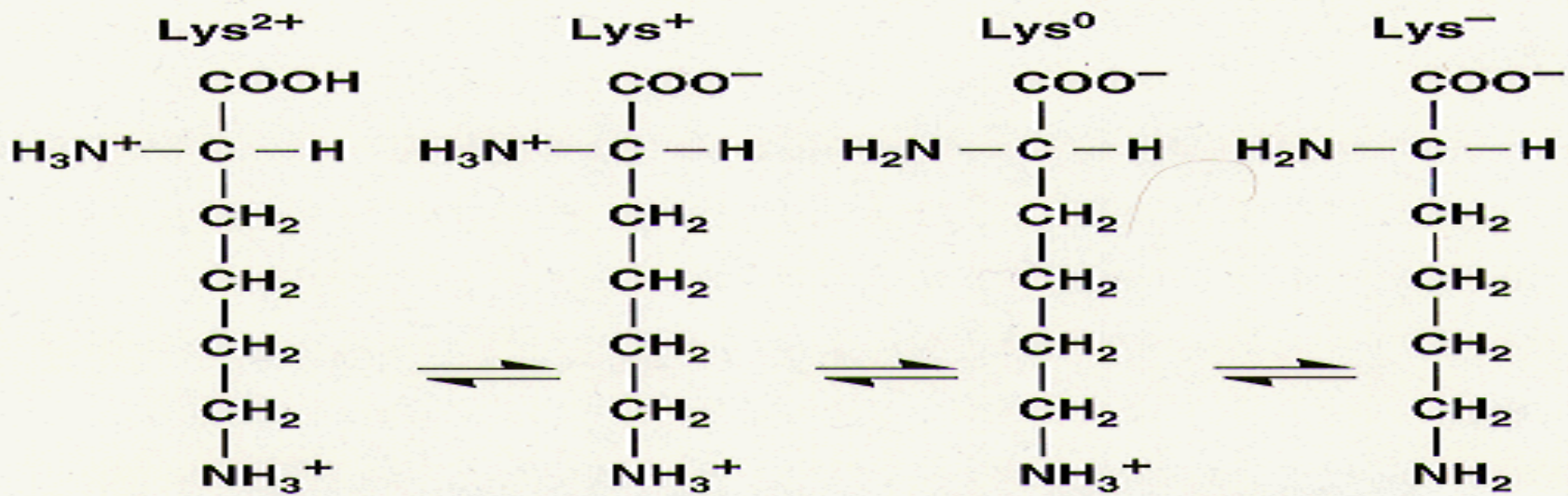
- Titration curve for glycine
 - Note that one starts with all groups in acid form.
 - Note how many equivalents are added
 - Note that at 0.5 and 1.5 equivalents, pH is equal to pK of group being titrated.
 - Note pH which gives zero charge is the isoelectric point. Calculated as $(\text{p}K_1 + \text{p}K_2)/2$
 - Note where the buffering capacity is best

- Acidic amino acids such as aspartic acid have a second carboxyl group that can donate and accept protons.
- The pI for aspartic acid occurs at a pH of 2.8.







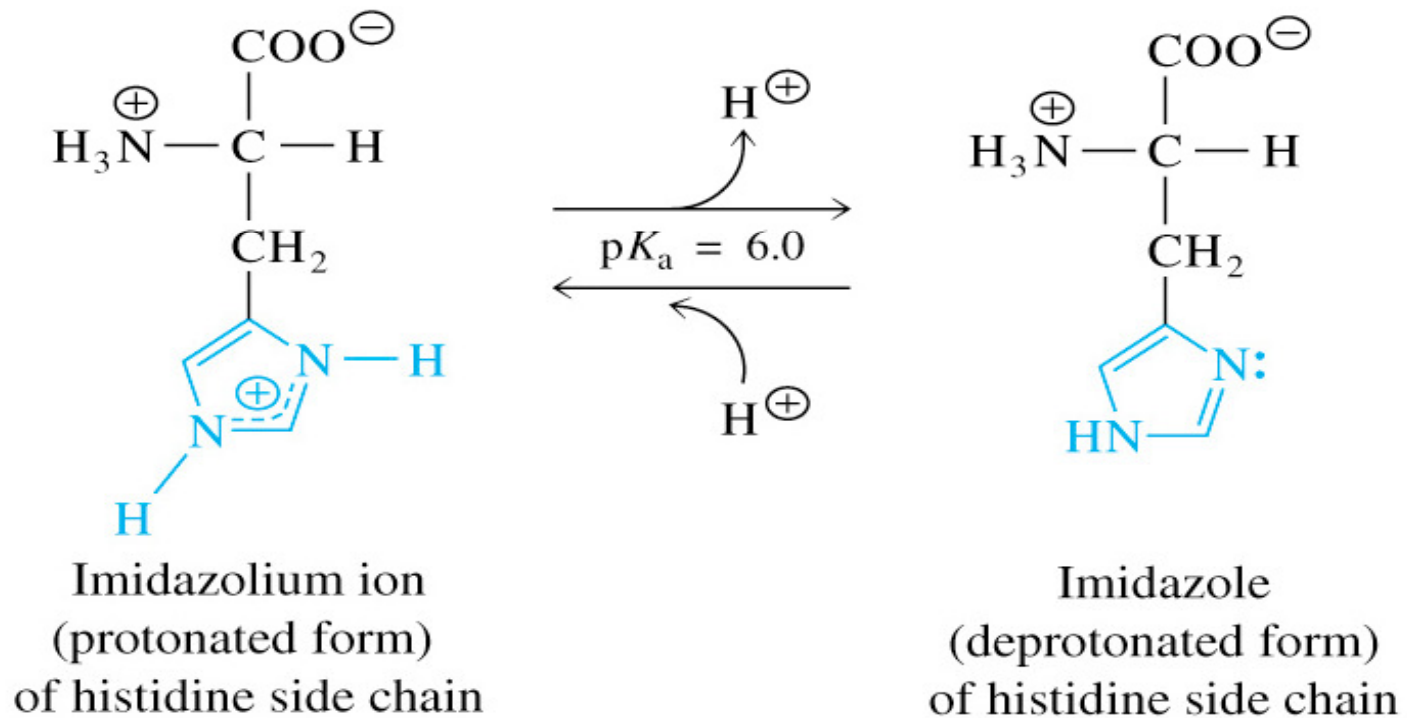


pK_a of amino acids

AA	COOH	NH ₃ ⁺	R	AA	COOH	NH ₃ ⁺	R
Ala	2.35	9.87	-	Leu	2.33	9.74	-
Arg	1.82	8.99	12.48	Lys	2.16	9.18	10.79
Asn	2.10	8.84	-	Met	2.13	9.28	-
Asp	1.99	9.90	3.90	Phe	2.16	9.18	-
Cys	1.92	10.78	8.33	Pro	1.95	10.65	-
Glu	2.10	9.47	4.07	Ser	2.19	9.21	~13
Gln	2.17	9.13	-	Thr	2.09	9.10	~13
Gly	2.35	9.78	-	Trp	2.43	9.44	-
His	1.80	9.33	6.04	Tyr	2.20	9.11	10.13
Ile	2.32	9.76	-	Val	2.29	9.74	-

Deprotonation of imidazolium ring

(b)



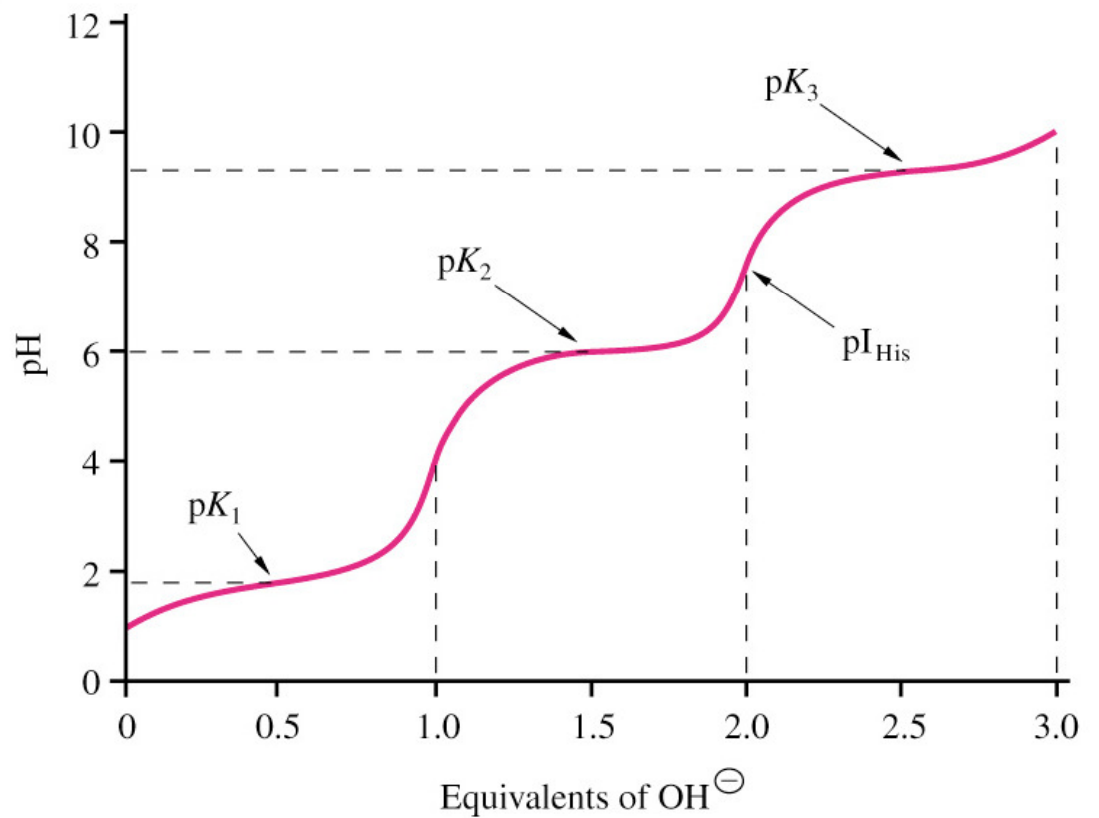
Ionization of Histidine

Titration curve (a)
of histidine

$$pK_1 = 1.8$$

$$pK_2 = 6.0$$

$$pK_3 = 9.3$$



- With a pK_a value near 6, the imidazole group can be uncharged or positively charged near neutral pH, depending on its local environment .
- histidine is often found in the active sites of enzymes, where the imidazole ring can bind and release protons in the course of enzymatic reactions.

- The imidazole ring of histidine allows it to act as either a proton donor or acceptor at physiological pH. Hence, it is frequently found in the reactive center of enzymes. Equally important is the ability of histidines in hemoglobin to buffer the H^+ ions from carbonic acid ionization in red blood cells. It is this property of hemoglobin that allows it to exchange O_2 and CO_2 at the tissues or lungs, respectively.

The Hydrophobicity of Amino Acid Side Chains

- **Hydropathy**: the relative hydrophobicity of each amino acid
- The larger the hydropathy, the greater the tendency of an amino acid to prefer a hydrophobic environment
- Hydropathy affects protein folding: hydrophobic side chains tend to be in the interior hydrophilic residues tend to be on the surface

- Hydropathy scale for amino acid residues
(Free-energy change for transfer of an amino acid from interior of a lipid bilayer to water)

Δ	Amino acid	Free-energy change for transfer (kJmol ⁻¹)
Highly hydrophobic		
	Isoleucine	3.1
	Phenylalanine	2.5
	Valine	2.3
	Leucine	2.2
	Methionine	1.1
Less hydrophobic		
	Tryptophan	1.5 ^b
	Alanine	1.0
	Glycine	0.67
	Cysteine	0.17
	Tyrosine	0.08
	Proline	20.29
	Threonine	20.75
	Serine	21.1
Highly hydrophilic		
	Histidine	21.7
	Glutamate	22.6
	Asparagine	22.7
	Glutamine	22.9
	Aspartate	23.0
	Lysine	24.6
	Arginine	27.5

Hydropathy Scale for Amino Acids

• Highly Hydrophobic	Less Hydrophobic	Highly Hydrophilic
Isoleucine	Tryptophan	Histidine
Valine	Alanine	Glutamic acid
Phenylalanine	Glycine	Asparagine
Leucine	Threonine	Lysine
Methionine	Serine	Arginine
	Proline	Aspartic acid
	Cysteine	
	Tyrosine	

Functional Significance of Amino Acid R-Groups

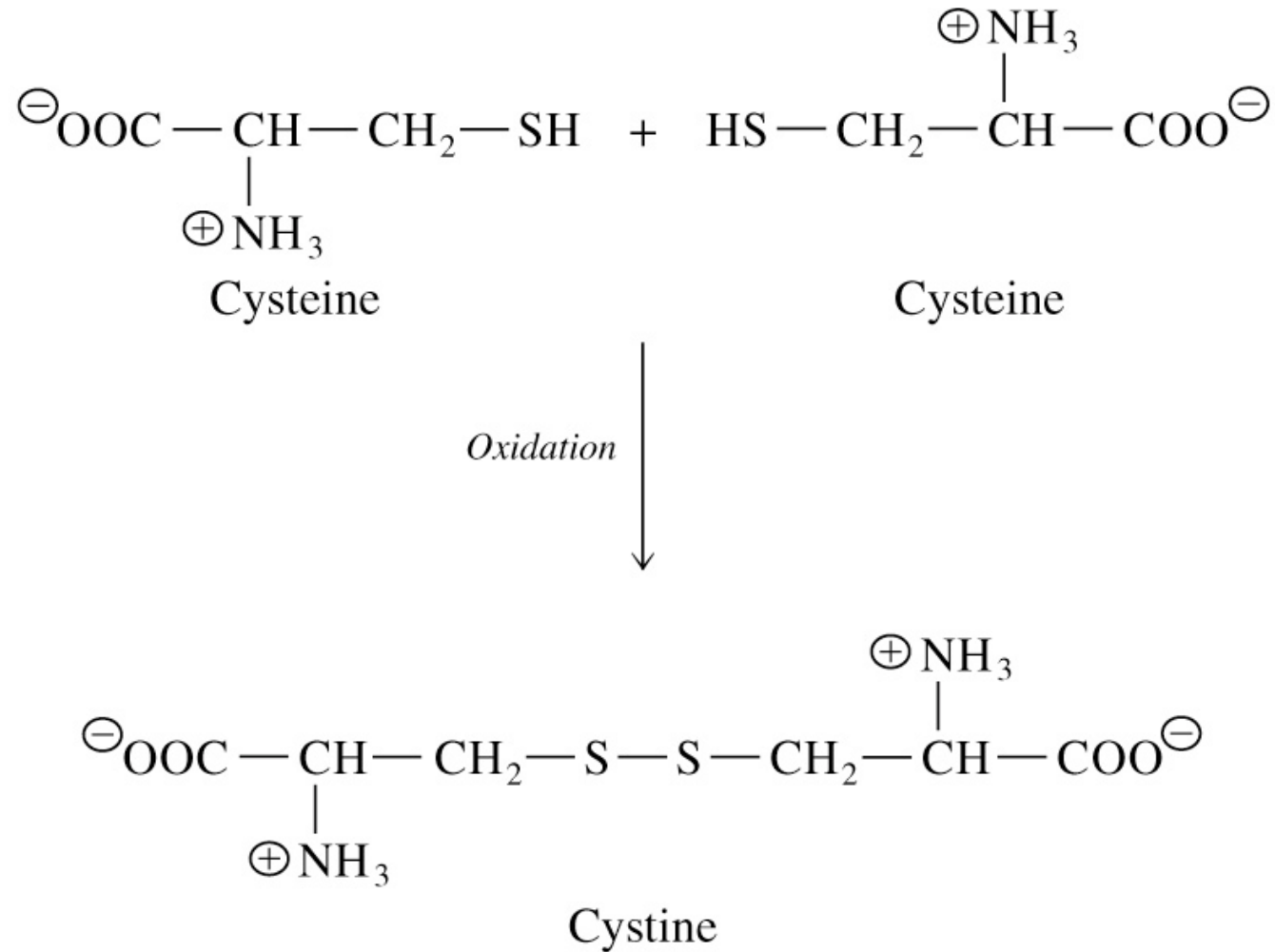
- * In solution it is the nature of the amino acid R-groups that dictate structure-function relationships of peptides and proteins.
 - * The hydrophobic amino acids will generally be encountered in the interior of proteins shielded from direct contact with water. Conversely, the hydrophilic amino acids are generally found on the exterior of proteins as well as in the active centers of enzymatically active proteins. The vary nature of certain amino acid R-groups that allow enzyme reactions to occur.

- * The larger aliphatic side chains are **hydrophobic** *that* is, they tend to cluster together rather than contact water.
- * The three-dimensional structures of water-soluble proteins are stabilized by this tendency of hydrophobic groups to come together, called **the hydrophobic effect** .
- * The different sizes and shapes of these hydrocarbon side chains enable them to pack together to form compact structures with few holes.
- * *Proline also has an aliphatic side chain, but it differs from other members of the set of 20 in that its side chain is bonded to both the nitrogen and the α -carbon atoms.*
- * Proline markedly influences protein architecture because its ring structure makes it more conformationally restricted than the other amino acids.

- *Proteins contain a wide range of functional groups. These functional groups include alcohols, thiols, thioethers,*
- carboxylic acids, carboxamides, and a variety of basic groups. When combined in various sequences, this array of
- functional groups accounts for the broad spectrum of protein function. For instance, the chemical reactivity associated
- with these groups is essential to the function of *enzymes, the proteins that catalyze specific chemical reactions in* biological systems

- The primary alcohol of serine and threonine as well as the thiol ($-SH$) of cysteine allow these amino acids to act as nucleophiles during enzymatic catalysis. Additionally, the thiol of cysteine is able to form a disulfide bond with other cysteines:

Formation of cystine



Disulfide-linked Cys residues

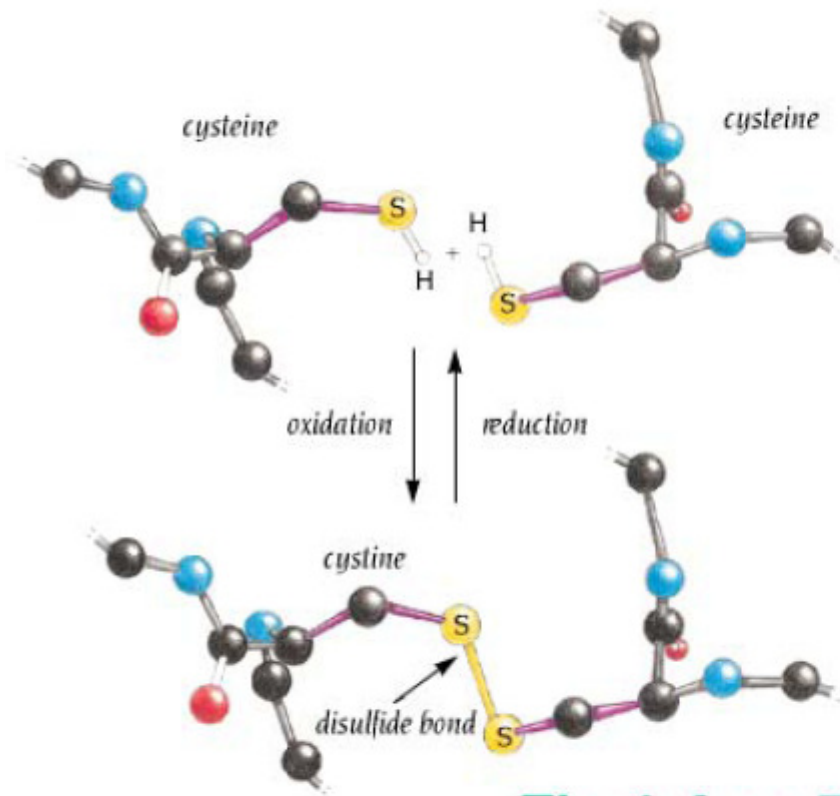
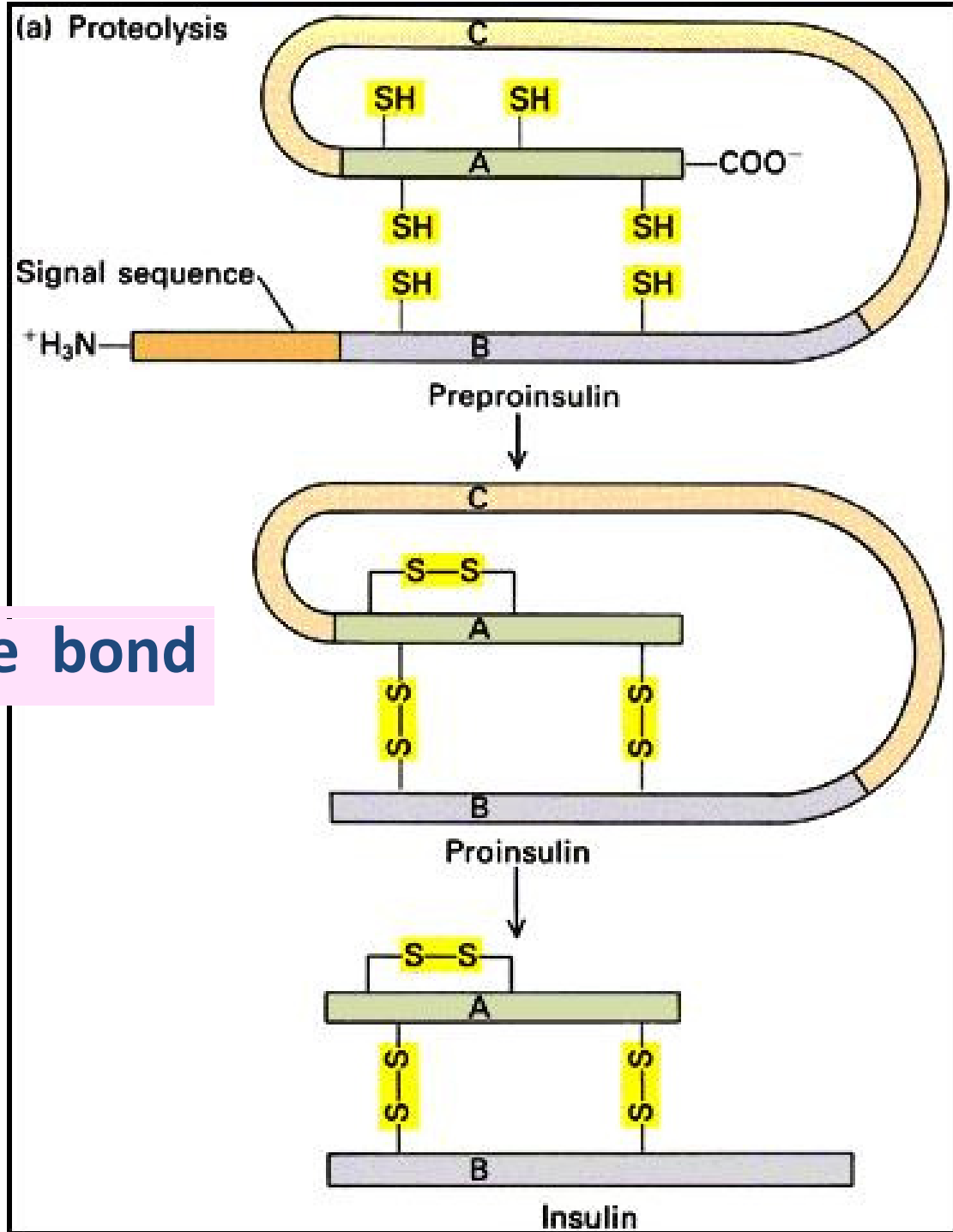


Fig 1.4, p. 5

- Two adjacent cysteine residues can be oxidized to form a disulfide bond
- disulfide bonds are usually found in extracellular and not intracellular proteins
 - inside of cell is a reducing environment
- disulfide bonds can stabilize protein structure by providing crosslink

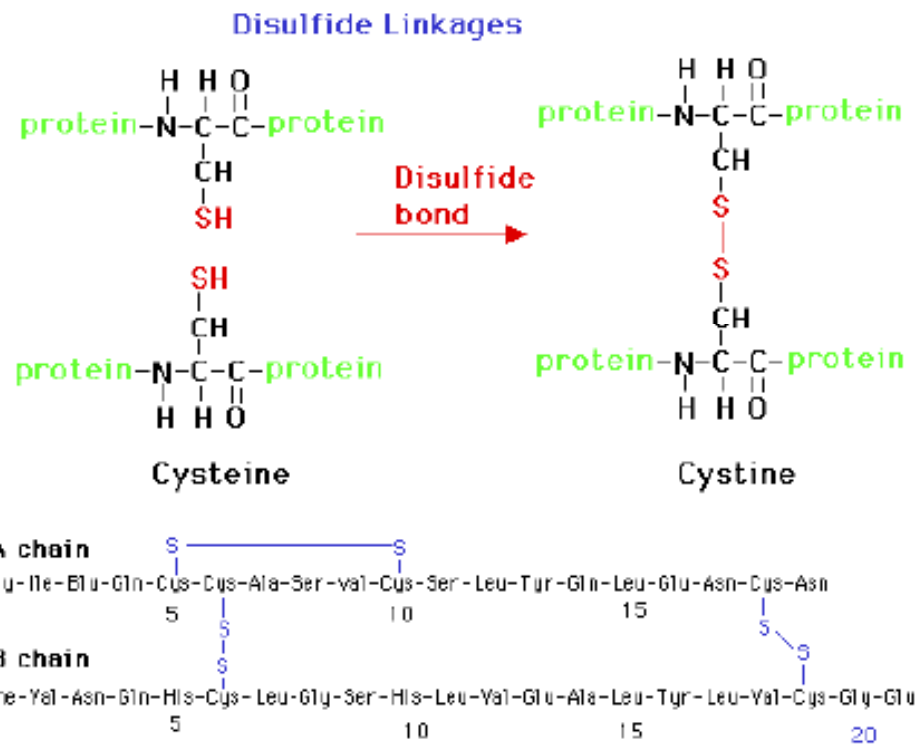
- This simple disulfide is identified as cystine. The formation of disulfide bonds between cysteines present within proteins is important to the formation of active structural domains in a large number of proteins. Disulfide bonding between cysteines in different polypeptide chains of oligomeric proteins plays a crucial role in ordering the structure of complex proteins, e.g. the insulin receptor.



Disulfide bond

Disulfide Bonds

- Pairs of cysteines can form disulfide bonds between different parts of the main chain
- This adds stability and is common in extracellular proteins



Properties and Conventions Associated with the Standard Amino Acids

Amino acid	Abbreviated names		M_r	pK_a values			pI	Hydropathy index*	Occurrence in proteins (%) [†]
				pK_1 (-COOH)	pK_2 (-NH ₃ ⁺)	pK_R (R group)			
Nonpolar, aliphatic R groups									
Glycine	Gly	G	75	2.34	9.60		5.97	-0.4	7.2
Alanine	Ala	A	89	2.34	9.69		6.01	1.8	7.8
Valine	Val	V	117	2.32	9.62		5.97	4.2	6.6
Leucine	Leu	L	131	2.36	9.60		5.98	3.8	9.1
Isoleucine	Ile	I	131	2.36	9.68		6.02	4.5	5.3
Methionine	Met	M	149	2.28	9.21		5.74	1.9	2.3
Aromatic R groups									
Phenylalanine	Phe	F	165	1.83	9.13		5.48	2.8	3.9
Tyrosine	Tyr	Y	181	2.20	9.11	10.07	5.66	-1.3	3.2
Tryptophan	Trp	W	204	2.38	9.39		5.89	-0.9	1.4
Polar, uncharged R groups									
Serine	Ser	S	105	2.21	9.15		5.68	-0.8	6.8
Proline	Pro	P	115	1.99	10.96		6.48	1.6	5.2
Threonine	Thr	T	119	2.11	9.62		5.87	-0.7	5.9
Cysteine	Cys	C	121	1.96	10.28	8.18	5.07	2.5	1.9
Asparagine	Asn	N	132	2.02	8.80		5.41	-3.5	4.3
Glutamine	Gln	Q	146	2.17	9.13		5.65	-3.5	4.2
Positively charged R groups									
Lysine	Lys	K	146	2.18	8.95	10.53	9.74	-3.9	5.9
Histidine	His	H	155	1.82	9.17	6.00	7.59	-3.2	2.3
Arginine	Arg	R	174	2.17	9.04	12.48	10.76	-4.5	5.1
Negatively charged R groups									
Aspartate	Asp	D	133	1.88	9.60	3.65	2.77	-3.5	5.3
Glutamate	Glu	E	147	2.19	9.67	4.25	3.22	-3.5	6.3

Amino acids (fully protonated)

