

CHAPTER 3

Amino Acids: Structure and Properties

Chapter at a Glance

The reader will be able to answer questions on the following topics:

- Classification of amino acids based on structure
- Based on side chain character
- Based on metabolic fate
- Based on nutritional requirements
- Isoelectric point
- Reactions due to carboxyl group
- Reactions due to amino group
- Reactions of SH group
- Peptide bond formation

Proteins are of paramount importance in biological systems. All the major structural and functional aspects of the body are carried out by protein molecules. All proteins are polymers of amino acids. Proteins are composed of a number of amino acids linked by peptide bonds.

Although about 300 amino acids occur in nature, only 20 of them are seen in human body. Most of the amino acids (except proline) are **alpha amino acids**, which means that the amino group is attached to the same carbon atom to which the carboxyl group is attached (Fig. 3.1).

CLASSIFICATION OF AMINO ACIDS

Based on Structure

A. Aliphatic amino acids

a. Monoamino monocarboxylic acids:

- Simple amino acids: Glycine, Alanine (Fig. 3.2)
- Branched chain amino acids: Valine, Leucine, Isoleucine (Fig. 3.3)
- Hydroxyamino acids: Serine, Threonine (Fig. 3.4.)
- Sulfur-containing amino acids: Cysteine, Methionine (Fig. 3.5)
- Amino acids with amide group: Asparagine, Glutamine (Fig. 3.6).

b. Monoamino dicarboxylic acids: Aspartic acid, Glutamic acid (Fig. 3.7).

c. Dibasic monocarboxylic acids: Lysine, Arginine (Fig. 3.8).

B. Aromatic amino acids:

Phenylalanine, Tyrosine (Fig. 3.9).

C. Heterocyclic amino acids:

Tryptophan (Fig. 3.10), Histidine (Fig. 3.11).

D. Imino acid: Proline (Fig. 3.11).

E. Derived amino acids:

- i. **Derived amino acids found in proteins:** After the synthesis of proteins, some of the amino acids are modified, e.g. hydroxy proline (Fig. 3.12) and hydroxy lysine are important components of collagen. Gamma carboxylation of glutamic acid residues of proteins is important for clotting process (Fig. 3.12). In ribosomal proteins and in histones, amino acids are extensively methylated and acetylated.
- ii. **Derived amino acids not seen in proteins** (Non-protein amino acids): Some derived amino acids are seen free in cells, e.g. Ornithine (Fig. 3.12), Citrulline, Homocysteine. These are produced during the metabolism of amino acids. Thyroxine may be considered as derived from tyrosine.
- iii. **Non-alpha amino acids:** Gamma amino butyric acid (GABA) is derived from glutamic acid. Beta alanine, where amino group is in beta position, is

a constituent of pantothenic acid (vitamin) and co-enzyme A.

Each amino acid will have three-letter and one-letter abbreviations which are shown in Table 3.1 as well as in Figures 3.2 to 3.11. Sometimes asparagine/aspartic acid may not be separately identified, for which 3-letter abbreviation is Asx and 1-letter abbreviation is B. Similarly Glx or Z stands for glutamine/glutamic acid.

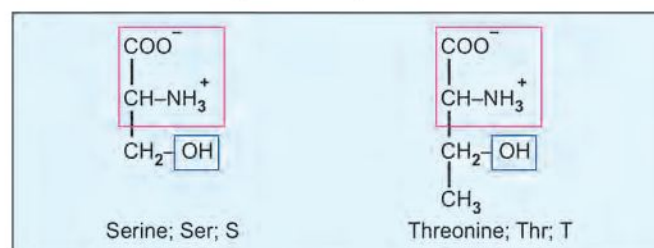


Fig. 3.4: Hydroxyamino acids



Fig. 3.5: Sulfur-containing amino acids



Fig. 3.6: Amino acids with amide groups



Fig. 3.7: Dicarboxylic amino acids

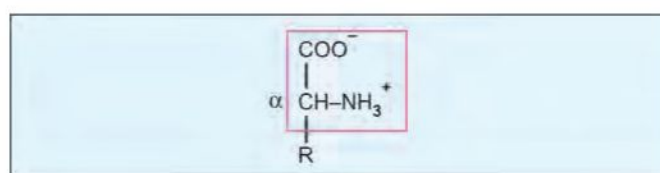


Fig. 3.1: General structure

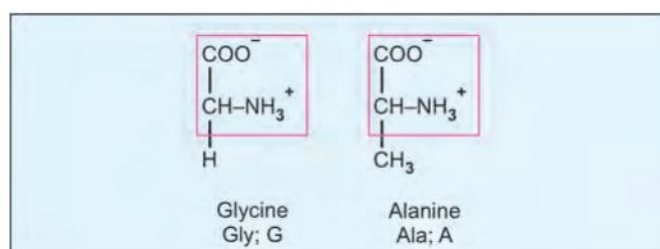


Fig. 3.2: Simple amino acids

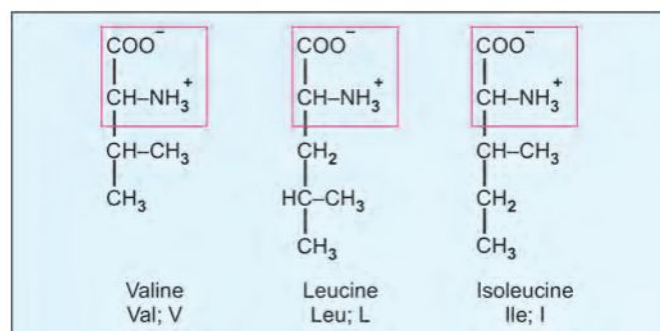


Fig. 3.3: Branched chain amino acids

Special Groups in Amino Acids

In the figures, special groups are shaded. Arginine contains **guanidinium** group; Phenylalanine (**benzene**); Tyrosine (**phenol**); Tryptophan (**indole**); Histidine (**imidazole**); and Proline (**pyrrolidine**) (Table 3.1). Proline has a secondary amino group, and hence it is an **imino acid**.

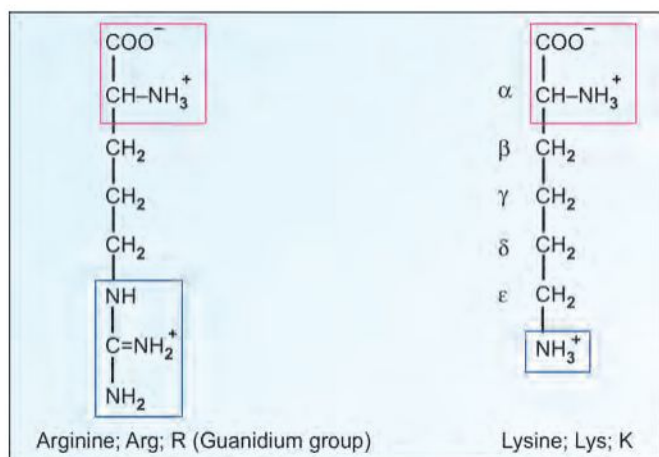


Fig. 3.8: Dibasic amino acids

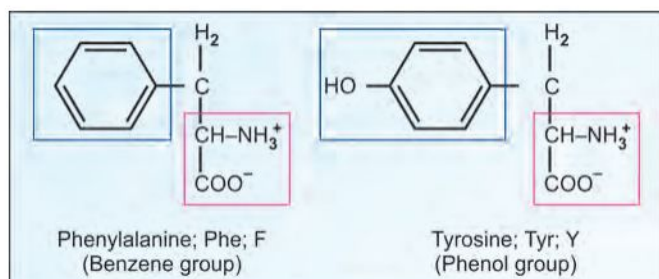


Fig. 3.9: Aromatic amino acids

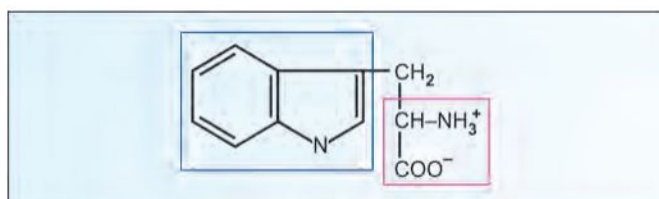


Fig. 3.10: Tryptophan (Trp) (W) with indole group

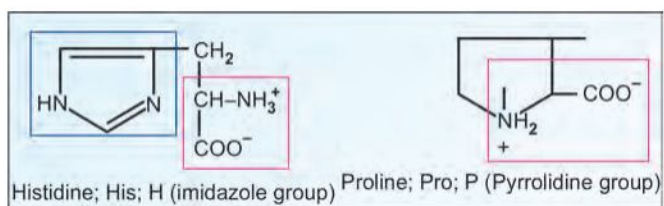


Fig. 3.11: Histidine and proline

Based on Side Chain

A. Amino acids having non-polar side chains:

These include Alanine, Valine, Leucine, Isoleucine, Methionine, Proline, Phenylalanine and Tryptophan. These groups are **hydrophobic** (water repellent) and lipophilic. Therefore, the parts of proteins made up of these amino acids will be hydrophobic in nature.

B. Amino acids having uncharged or non-ionic polar side chains:

Glycine, Serine, Threonine, Cysteine, Tyrosine, Glutamine and Asparagine belong to this group. These amino acids are **hydrophilic** in nature.

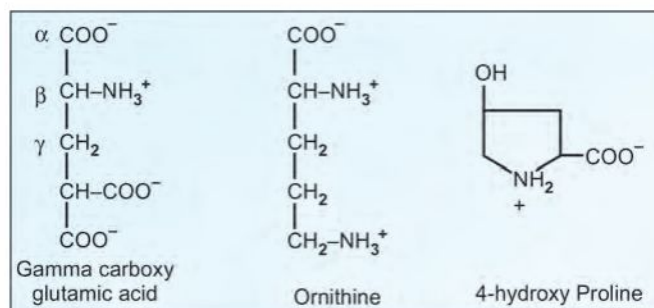


Fig. 3.12: Some derived amino acids

TABLE 3.1: Common amino acids

Name of amino acid	Special group present	3-letter abbreviation	1-letter abbreviation	Molecular weight
Glycine		Gly	G	77
Alanine		Ala	A	89
Valine		Val	V	117
Leucine		Leu	L	131
Isoleucine		Ile	I	131
Serine	Hydroxyl	Ser	S	105
Threonine	Hydroxyl	Thr	T	119
Cysteine	Sulfhydryl	Cys	C	121
Methionine	Thioether	Met	M	149
Asparagine	Amide	Asn	N	132
Glutamine	Amide	Gln	Q	146
Aspartic acid	β -carboxyl	Asp	D	133
Glutamic acid	γ -carboxyl	Glu	E	147
Lysine	ϵ -amino	Lys	K	146
Arginine	Guanidinium	Arg	R	174
Phenylalanine	Benzene	Phe	F	165
Tyrosine	Phenol	Tyr	Y	181
Tryptophan	Indole	Trp	W	204
Histidine	Imidazole	His	H	155
Proline (imino acid)	Pyrrolidine	Pro	P	115

(Tyrosine and Cysteine may show hydrophobic character when present in the interior of the protein).

C. Amino acids having charged or ionic polar side chains (hydrophilic):

- Acidic amino acids:** They have a negative charge on the R group: Aspartic acid and Glutamic acid. (Tyrosine is mildly acidic).
- Basic amino acids:** They have a positive charge on the R group: Lysine, Arginine and Histidine.

Based on Metabolism

- Purely ketogenic:** Leucine is purely ketogenic because it is converted to ketone bodies (see Fig.18.16).
- Ketogenic and glucogenic:** Lysine, Isoleucine, Phenylalanine, Tyrosine and Tryptophan are partially ketogenic and partially glucogenic. During metabolism, part of the carbon skeleton of these amino acids will enter the ketogenic pathway and the other part to glucogenic pathway (see Fig.18.16).
- Purely glucogenic:** All the remaining 14 amino acids are purely glucogenic as they enter only into the glucogenic pathway (see Chapter 18).

Based on Nutritional Requirements

- Essential or indispensable:** The amino acids may further be classified according to their essential nature for growth. Thus, **Isoleucine, Leucine, Threonine, Lysine, Methionine, Phenylalanine, Tryptophan, and Valine** are essential amino acids. Their carbon skeleton cannot be synthesized by human beings and so preformed amino acids are to be taken in food for normal growth. Normal growth and optimal health will not occur, if one such amino acid is deficient in the diet. See memory aid in Box 3.1.
- Partially essential or Semiessential: Histidine and Arginine** are semi-indispensable amino acids

Growing children require them in food. But they are not essential for the adult individual.

- Non-essential or Dispensable:** The remaining 10 amino acids are non-essential, because their carbon skeleton **can be synthesized** by the body. So we need not have to ingest these amino acids as such. However, they are also required for normal protein synthesis. The non-essential amino acids are Alanine, Asparagine, Aspartic acid, Cysteine, Glutamine, Glutamic Acid, Glycine, Proline, Serine and Tyrosine. All body proteins do contain all the non-essential amino acids.
- Conditionally essential amino acids:** When a person is suffering from a moderate to severe chronic illness, person may lose the ability to manufacture enough non-essential amino acids and thus require supplementation. Problems with digestion will also necessitate supplementation of “non-essential” amino acids. These amino acids are normally non-essential, but become essential during times of physiological stress. Then these amino acids have to be taken in food or through supplements. These conditionally essential amino acids are Arginine, Glycine, Cysteine, Tyrosine, Proline, Glutamine and Taurine.

Naming (numbering) of Carbon Atoms

Carbon atoms in amino acids in sequence are named with letters of Greek alphabets, starting from the carbon atom to which carboxyl group is attached. As examples, naming of glutamic acid is shown in Figure in 3.7 and that of lysine is shown in Figure 3.8.

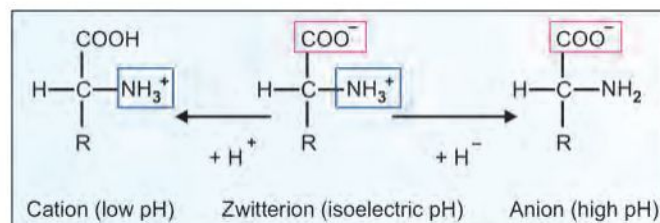


Fig. 3.13: Ionic forms of amino acids

GENERAL REACTIONS OF AMINO ACIDS

Due to Carboxyl Group

Decarboxylation

The amino acids will undergo alpha decarboxylation to form the corresponding amine (Fig. 3.17). Thus, some important amines are produced from amino acids. For example,

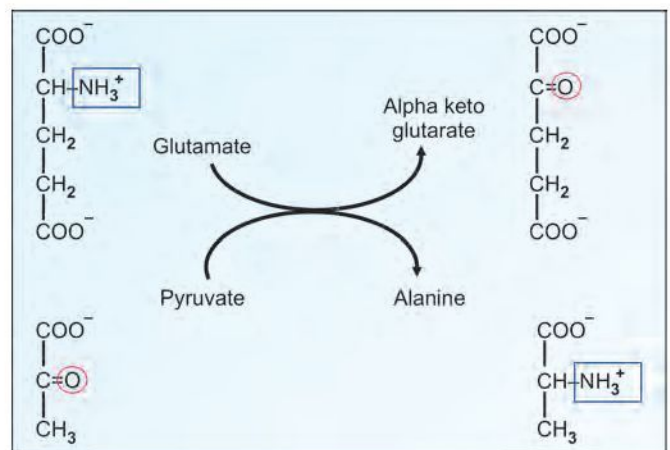
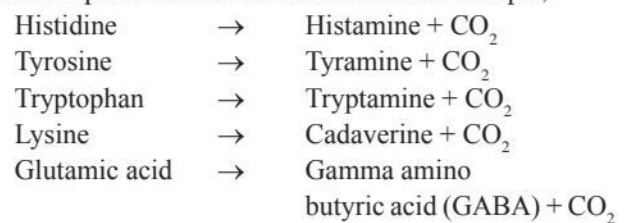


Fig. 3.18: Transamination reaction

Amide Formation

The -COOH group of dicarboxylic amino acids (other than alpha carboxyl) can combine with ammonia to form the corresponding amide. For example,



These amides are also components of protein structure. The amide group of glutamine serves as the source of nitrogen for nucleic acid synthesis.

Due to Amino Group

Transamination

The alpha amino group of amino acid can be transferred to alpha keto acid to form the corresponding new amino acid and alpha keto acid (Fig. 3.18). This is an important reaction in the body for the interconversion of amino acids and for **synthesis of non-essential amino acids**.

Oxidative Deamination

The alpha amino group is removed from the amino acid to form the corresponding keto acid and ammonia (see Fig. 14.9). In the body, **Glutamic acid** is the most common amino acid to undergo oxidative deamination.

Formation of Carbamino Compound

Carbon dioxide adds to the alpha amino group of amino acids to form carbamino compounds. The reaction occurs at alkaline pH and serves as a mechanism for the transport of carbon dioxide from tissues to the lungs by hemoglobin (see Chapter 22).

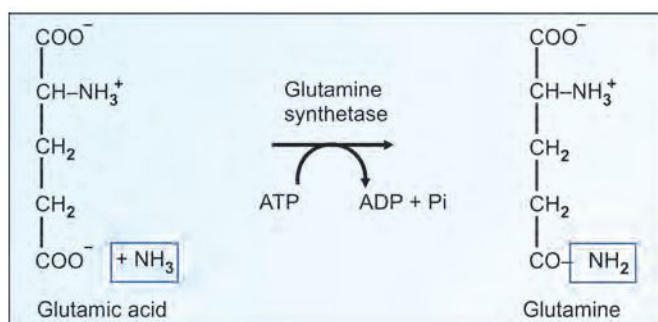
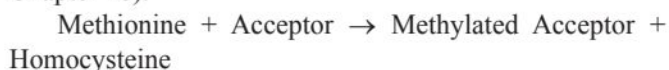


Fig. 3.19: Formation of glutamine

Due to Side Chains

Transmethylation

The methyl group of Methionine, after activation, may be transferred to an acceptor, which becomes methylated (see Chapter 16).



Ester Formation by the OH Group

The hydroxy amino acids can form esters with phosphoric acid. In this manner, the **Serine and Threonine** residues of proteins are involved in the formation of phosphoproteins. Similarly, these hydroxyl groups can form O-glycosidic bonds with carbohydrate residues to form glycoproteins.

Reaction of the Amide Group

The amide groups of **Glutamine and Asparagine** can form N-glycosidic bonds with carbohydrate residues to form glycoproteins.

Reactions of SH Group

Cysteine has a sulfhydryl (SH) group and it can form a disulfide (S-S) bond with another cysteine residue. The two cysteine residues can connect two polypeptide chains by the formation of **interchain disulfide bonds** or links (Fig. 3.20). The dimer formed by two cysteine residues is sometimes called **Cystine** or Dicysteine.

Amino Acid Derivatives of Importance

- Gamma amino butyric acid (GABA)**, a derivative of glutamic acid) and dopamine (derived from tyrosine)

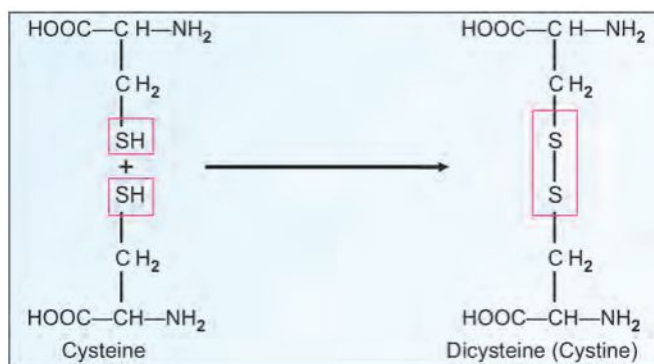


Fig. 3.20: Formation of disulfide bridges

are neurotransmitters. **GABA pentin** can pass blood brain barrier and can form GABA in brain.

- ii. **Histamine** (synthesized from histidine) is the mediator of allergic reactions.
- iii. **Thyroxine** (from tyrosine) is an important thyroid hormone.
- iv. **Cycloserine**, a derivative of serine is an antituberculous drug. **Azaserine** inhibits reactions where amide groups are added, and so acts as an anticancer drug.
- v. **Histidine** residues are important in the buffering activity of proteins.
- vi. **Ornithine** and **citrulline** are derivatives of arginine, and are essential for urea synthesis.

PEPTIDE BOND FORMATION

Alpha carboxyl group of one amino acid reacts with **alpha amino group** of another amino acid to form a peptide bond or CO-NH bridge (Fig. 3.21). Proteins are made by polymerization of amino acids through peptide bonds. Details of protein structures are given in Chapter 4.

Color Reactions of Amino Acids and Proteins

1. Ninhydrin Reaction (Ruhemann, 1910)

Amino acid + 2 molecules of ninhydrin $\xrightarrow{\text{heat}}$ Aldehyde with one carbon atom less + CO_2 + purple complex.

All amino acids when heated with ninhydrin can form complexes; pink, purple or blue in color. The color complex is called **Ruhemann's purple**. Proline and hydroxy proline will give yellow color with ninhydrin. Amino acids with amide group (glutamine, asparagine) produce a brown color. The ninhydrin reaction may be adopted for qualitative as well as quantitative estimation of amino acids. It is often used for detection of amino acids in chromatography. Proteins do not give a true color reaction;

but N-terminal end amino group of protein will also react with ninhydrin, to produce a blue color.

2. **Biuret Reaction** : Cupric ions in an alkaline medium form a violet color with peptide bond nitrogen (Schiff, 1896). This needs a minimum of two **peptide bonds**, and so *individual amino acids and dipeptides will not answer this test*. This reaction can be used for quantitative estimation also. The name is derived from the compound biuret ($\text{NH}_2\text{—CO—NH—CO—NH}_2$), a condensation product of two urea molecules, which also gives a positive color test. Magnesium and ammonium sulfates interfere with this reaction.
3. **Xanthoproteic Test**: The ring systems in **phenylalanine, tyrosine and tryptophan** undergo nitration on treatment with concentrated nitric acid when heated (Salkowski, 1888). The end product is yellow in color which is intensified in strong alkaline medium. This reaction causes the yellow stain in skin by nitric acid.
4. **Millon's Test**: The phenol group of **phenylalanine and tyrosine** containing proteins, when heated with mercuric sulfate in sulfuric acid and sodium nitrite (or, mercurous and mercuric nitrates in nitric acid) form red colored mercury phenolate (Millon, 1812–1867). Chloride interferes with this reaction and so it is not suitable to test for tyrosine in urine samples. Both xanthoproteic and Millon's tests are negative for tapioca (casava), which is deficient in phenylalanine and tyrosine.
5. **Aldehyde Tests for Tryptophan**: In the **Hopkins-Cole test**, tryptophan containing protein is mixed with glyoxylic acid, and the mixture is layered over concentrated sulfuric acid. A violet ring at the interface of liquids shows the presence of the indole ring.

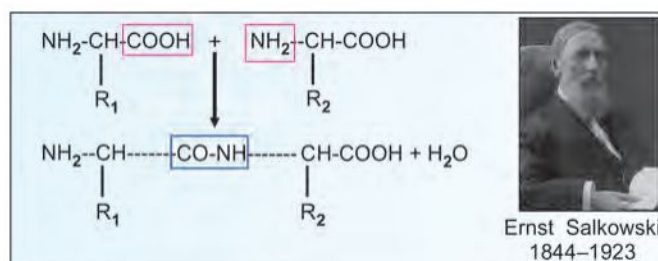


Fig. 3.21: Peptide bond formation

TABLE 3.3: Color reactions of amino acids

Reaction	Answered by specific group
1. Ninhydrin	Alpha amino group
2. Biuret reaction	Peptide bonds
3. Xanthoproteic test	Benzene ring (Phe, Tyr, Trp)
4. Millon's test	Phenol (Tyrosine)
5. Aldehyde test	Indole (Tryptophan)
6. Sakaguchi's test	Guanidinium (Arginine)
7. Sulfur test	Sulfhydryl (Cysteine)
8. Nitroprusside test	Sulfhydryl (Cysteine)
9. Pauly's test	Imidazole (Histidine)

Box 3.2: Selenocysteine as the 21st amino acid

21st century witnesses the addition of **Selenocysteine** as the 21st amino acid present in human proteins. An amino acid is given the individual status, when it is incorporated as such into proteins during protein biosynthesis, and having a separate codon. Selenocysteine is present in some enzymes. Instead of SH (sulfhydryl) group in cysteine, SeH (selenium) is present in selenocysteine. It is abbreviated as SeCys or SeC. Details are given in Chapter 16, under serine. There are about 25 proteins that incorporate selenocysteine.

Similarly **Pyrrolysine** (Pyl) is known as the 22nd amino acid. Pyrrolysine is a lysine in an amide linkage to substituted-pyrroline-5-carboxylate. It is present in methyl transferase enzymes of certain bacteria. Both SeC and Pyl are encoded by codons that normally function as stop signals.

Formaldehyde and mercuric sulfate is used similarly in **Acree-Rosenheim reaction** to get a violet color. Para-dimethyl-amino-benzaldehyde and strong hydrochloric acid give dark blue color (Ehrlich's reaction). Gelatin with limited tryptophan content will not answer these tests.

6. **Sakaguchi's Test for Arginine** : Free arginine or arginyl residues in proteins react with alpha-naphthol and alkaline hypobromite to give bright-red color. This is due to the guanidium group.
7. **Sulfur Test for Cysteine**: When cysteine or cysteine-containing proteins are boiled with strong alkali, organic sulfur splits and forms sodium sulfide, which on addition of lead acetate produces lead sulphide as a black precipitate. Methionine does not answer this test because sulfur in methionine is in the thioether linkage which is difficult to break. Albumin and keratin will answer sulfur test positively; but casein will give a negative test.
8. **Nitroprusside Reaction for SH groups**: Proteins with free sulfhydryl groups give a reddish color with sodium nitroprusside, in ammoniacal solution. Many proteins give a negative, reaction in the native state, but when denatured, reaction will be positive, showing the emergence of free SH groups.
9. **Pauly's Test for Histidine or Tyrosine**: Diazo-benzene sulfonic acid reacts with imidazole group of Histidine to form a cherry-red colored diazotized product under alkaline conditions. The same reagent will give an orange red colored product with phenol group of Tyrosine.

Color reactions of amino acids are shown as summary in Table 3.3. The amino acid **Selenocysteine** is described

in Box 3.2. Quantitative estimation procedures of proteins are given in Chapter 4.

QUICK LOOK OF CHAPTER 3

1. Most amino acids in the body are alpha amino acids.
2. Amino acids can be classified based on their (i) Structure (ii) Side chain characters (iii) Metabolic fate (iv) Nutritional requirements.
3. In solution, amino acids exist as 'Zwitter ions' or 'Ampholytes' at their characteristic Isoelectric pH (pI). In this state they carry no net charge.
4. The Imidazolium group of Histidine has a pK of 6.1 and contributes to the buffering actions of plasma proteins and hemoglobin at the physiological pH.
5. Glycine has no asymmetric carbon atoms and therefore has no optical activity.
6. Alpha carboxyl group of one amino acid combines with the alpha amino group of another amino acid to form a peptide bond. A peptide bond is a partial bond and hence there is no freedom of rotation.
7. Proteins are polymers of amino acids linked adjacently by peptide bonds.

Importance of side chains of amino acids:

α carboxy groups and a amino groups in proteins are unavailable as they form peptide bonds; nature of side chains determine physical properties, including protein folding.

Non-polar side chain

Alanine
Glycine
Isoleucine
Leucine
Methionine
Phenylalanine
Proline
Tryptophan
Valine

Uncharged polar side chains

Asparagine
Cysteine
Glutamine
Serine
Threonine
Tyrosine

Acidic side chains

Aspartic acid
Glutamic acid

Basic side chains

Arginine
Histidine
Lysine

Found on the outside of proteins that function in an aqueous environment and in the interior of membrane-associated proteins

Found in the interior of proteins that function in an aqueous environment and on the surface of proteins (such as membrane proteins) that interact with lipids



TOTOZ
— COLLECTION —