AMINO ACID
VERSUS
PROTEIN

AMINO ACID	PROTEIN
An amino acid refers to a simple organic molecule, containing both carboxyl and amino group	A protein refers to a large, nitrogenous, organic compound, composed of one or two amino acid chains
Building blocks of proteins	String of amino acids
Only some can be synthesized inside the body	All can be synthesized inside the body
Have a small molecular mass	Have a higher moecular mass
Twenty amino acids are involved in the synthesis of proteins	More than 10 million of proteins are produced in biological organisms
Composed of an amino group (NH2), a carboxylic acid group (COOH), a hydrogen atom (H), and a variable side chain (R)	Made up of amino acids linked together by peptide bonds
Involved in the protein synthesis	Serve as a structural, functional, and regulatory molecule in the body

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Classification of amino acids

Amino acid classification based on the structure

The 20 standard amino acids found in protein structure are divided into seven distinct groups.

1.Aliphatic amino acids:

2.Hydroxyl group containing amino acids:

3.Sulfur containing amino acids:

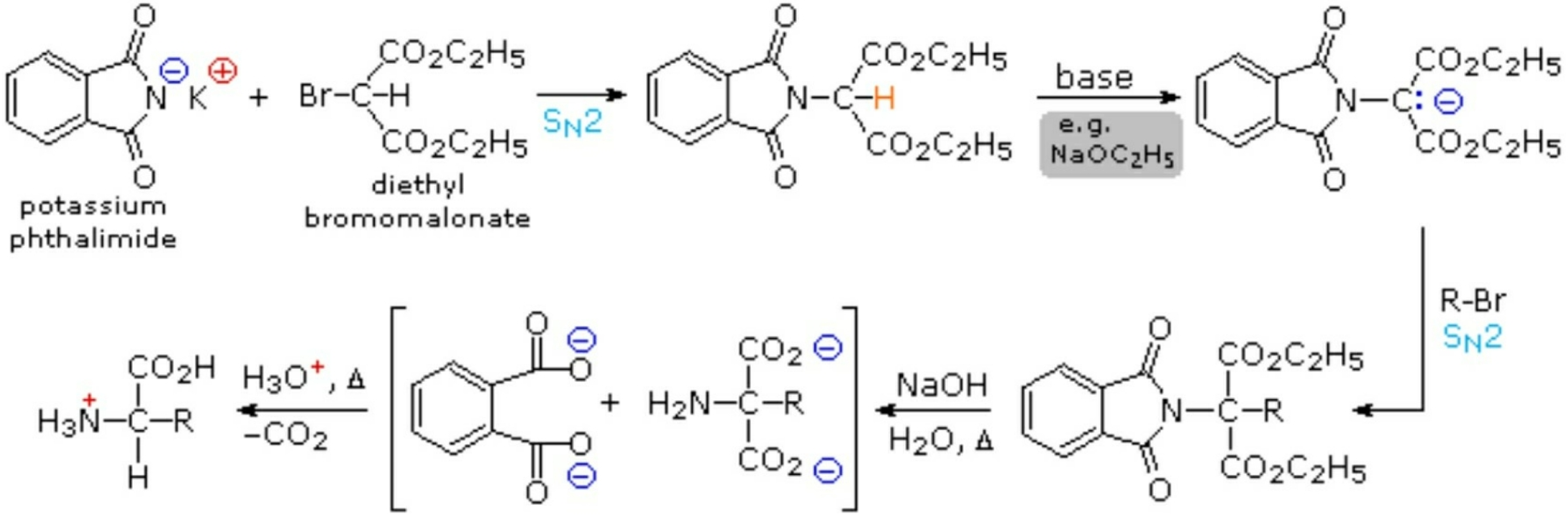
4.Acidic amino acids and their amides:

5.Basic amino acids:

6.Aromatic amino acids:

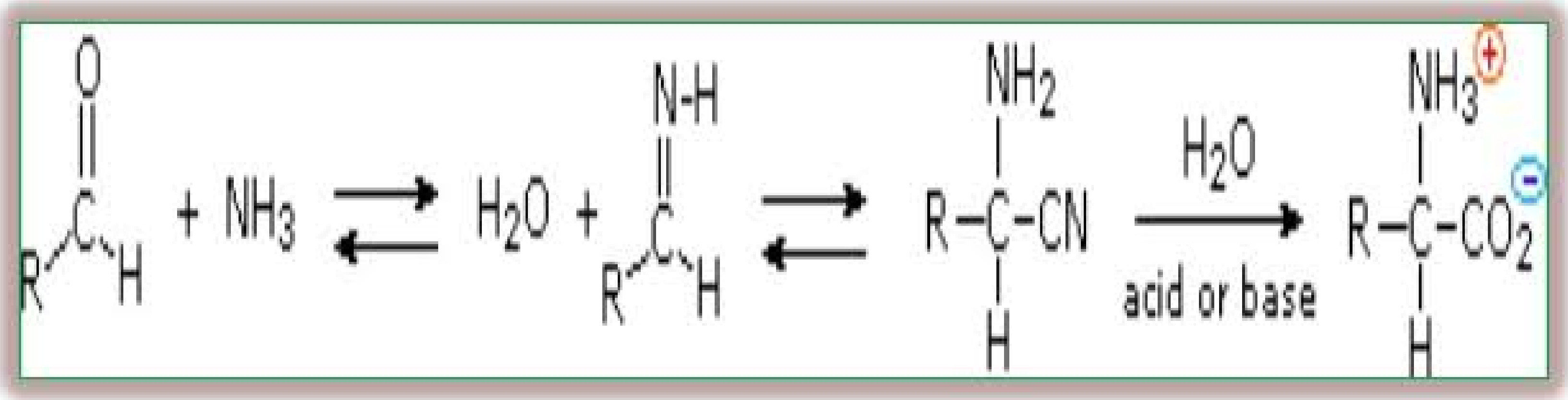
7.Imino acids:

Gabriel Phthalimide synthesis.



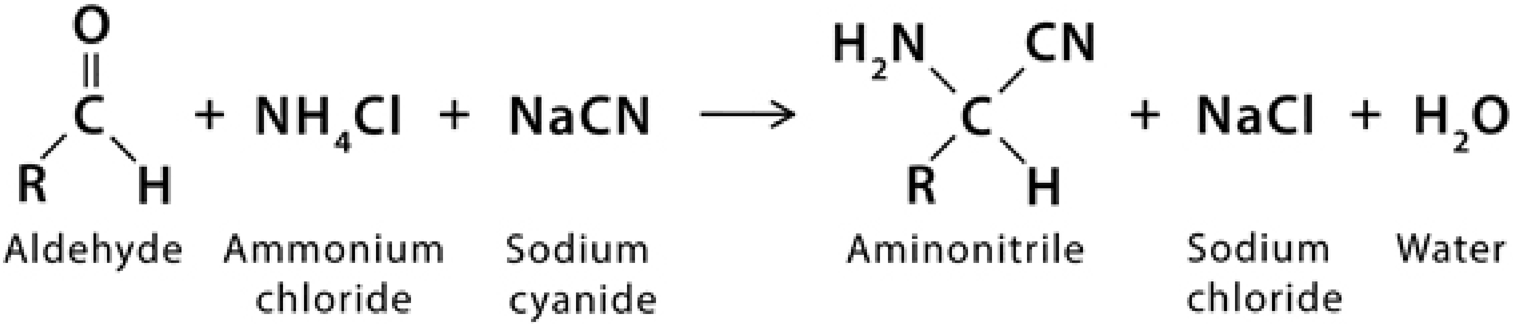
3- Strecker Synthesis

- assembles an alpha-amino acid from ammonia (the amine precursor), cyanide (the carboxyl precursor) and an aldehyde. This reaction is essentially an imino analog of cyanohydrin formation. The alpha-amino nitrile formed in this way can then be hydrolyzed to an amino acid by either acid or base catalysis.

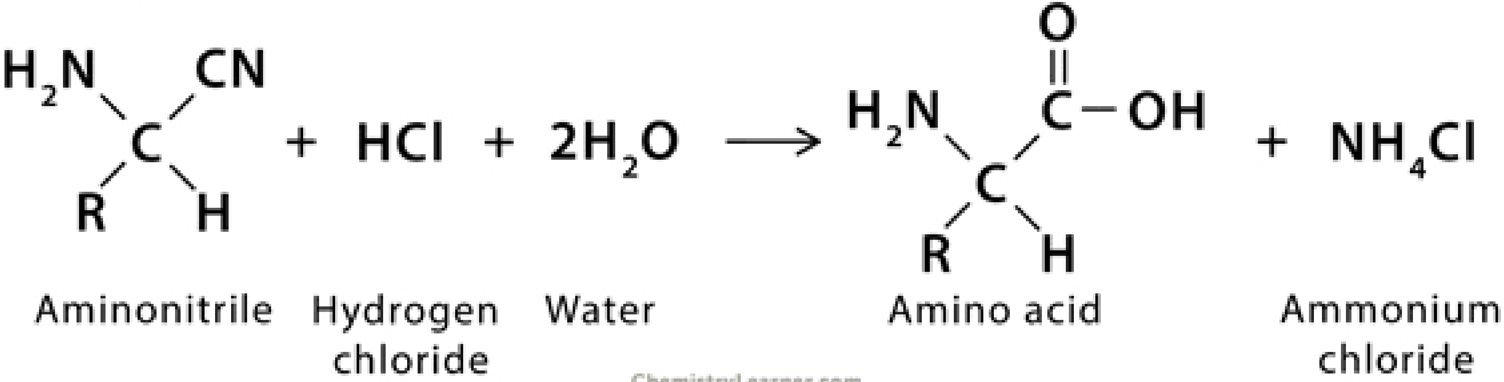


Strecker Synthesis

Step 1: Formation of aminonitrile

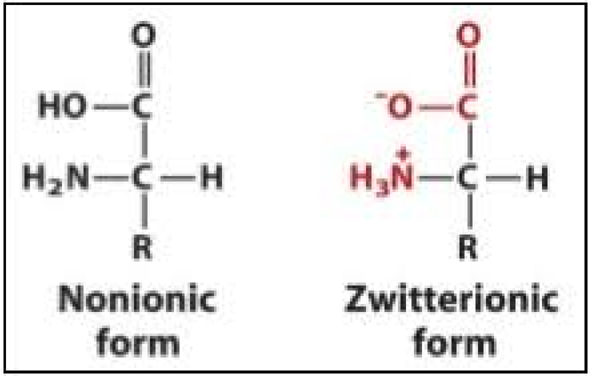


Step 2: Hydrolysis of aminonitrile to yield amino acid



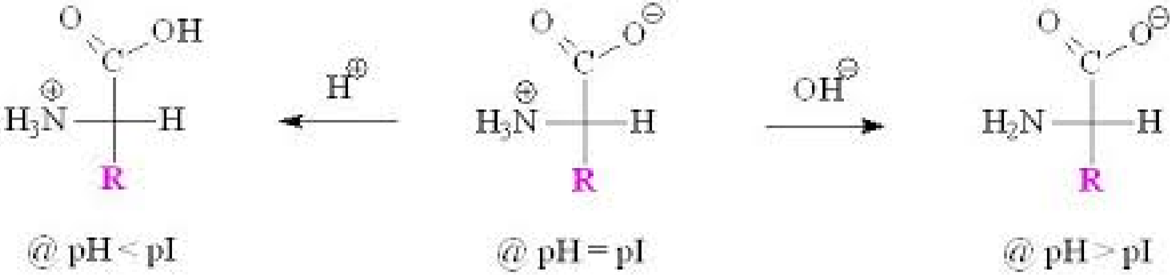
Zwitterions

- Although α-amino acids are commonly written in the unionized form, they are more properly written in the zwitterion (internal salt) form (Germ. Zwitter means hybrid)
- Both the $-NH_2$ and the $-COOH$ groups in an amino acid undergo ionization in water.
- At physiological pH (7.4), a zwitterion forms
- Both + and − charges
- Overall neutral
- Amphoteric
    - Amino group is protonated
    - Carboxyl group is deprotonated
    - **Soluble** in polar solvents due to ionic character



Isoelectric Point

- Each amino acid has an **isoelectric point**, (**pI**) numerically equal to the pH at which the **zwitterion** concentration is at a maximum.
- The amino acid has no NET charge at its pI; it has one positive and one negative charge.
- At a pH less than the value of the isoelectric point, the amino acid is protonated and has a POSITIVE charge; at a pH greater than the pI the amino acid is deprotonated and has a NEGATIVE charge.



Cation **Neutral** **Anion**

(zwitterion form)

End-group Analysis

- Number of chains can be determine by identifying the number of N- and C-terminal.
- N-terminal analysis
  - Dansyl chloride or FDNB method
  - Phenylisothiocynate (PITC)/ Edman reagent
  - Aminopeptidase
- C-terminal analysis
  - carboxypeptidase

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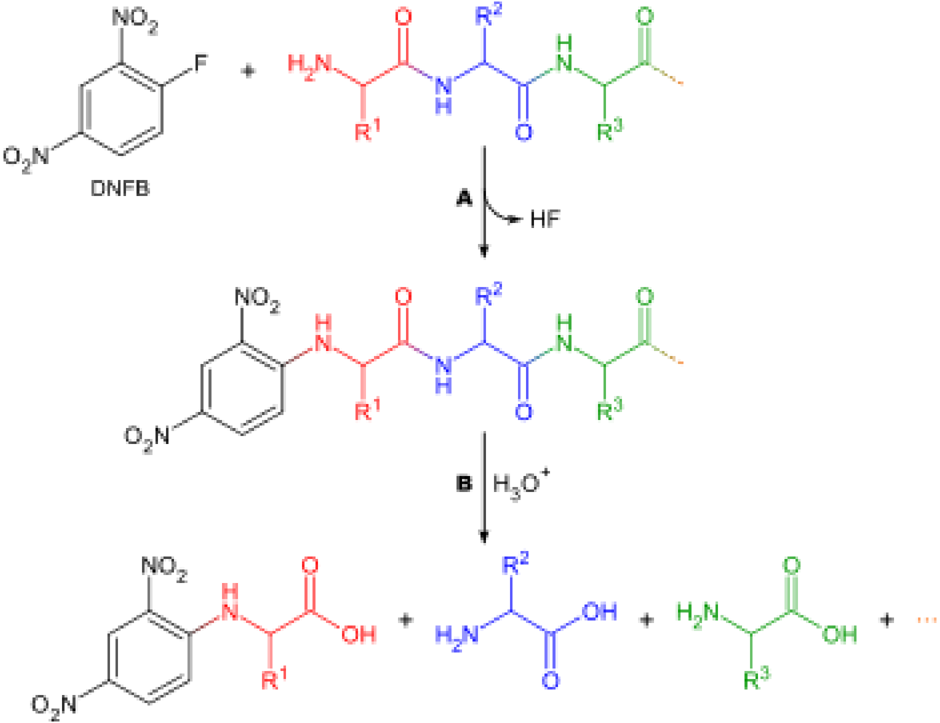
END GROUP ANALYSIS:-

❖ **N-terminal residue analysis**:-

❑ ***Sanger's Method:-***

➢ The peptide is treated with 2,4-dinitrofluorobenzene (DNFB) in presence of mildly basic solution of aqueous sodium bicarbonate at room temperature.

➢ Acid hydrolysis gives the DNP derivative of the N-terminal amino acid which is extracted and identified by chromatography and comparison with the standards.


NO2
F
O2N
DNFB
+
H2N
R1
R2
R3
A
HF
B
H3O+
OH

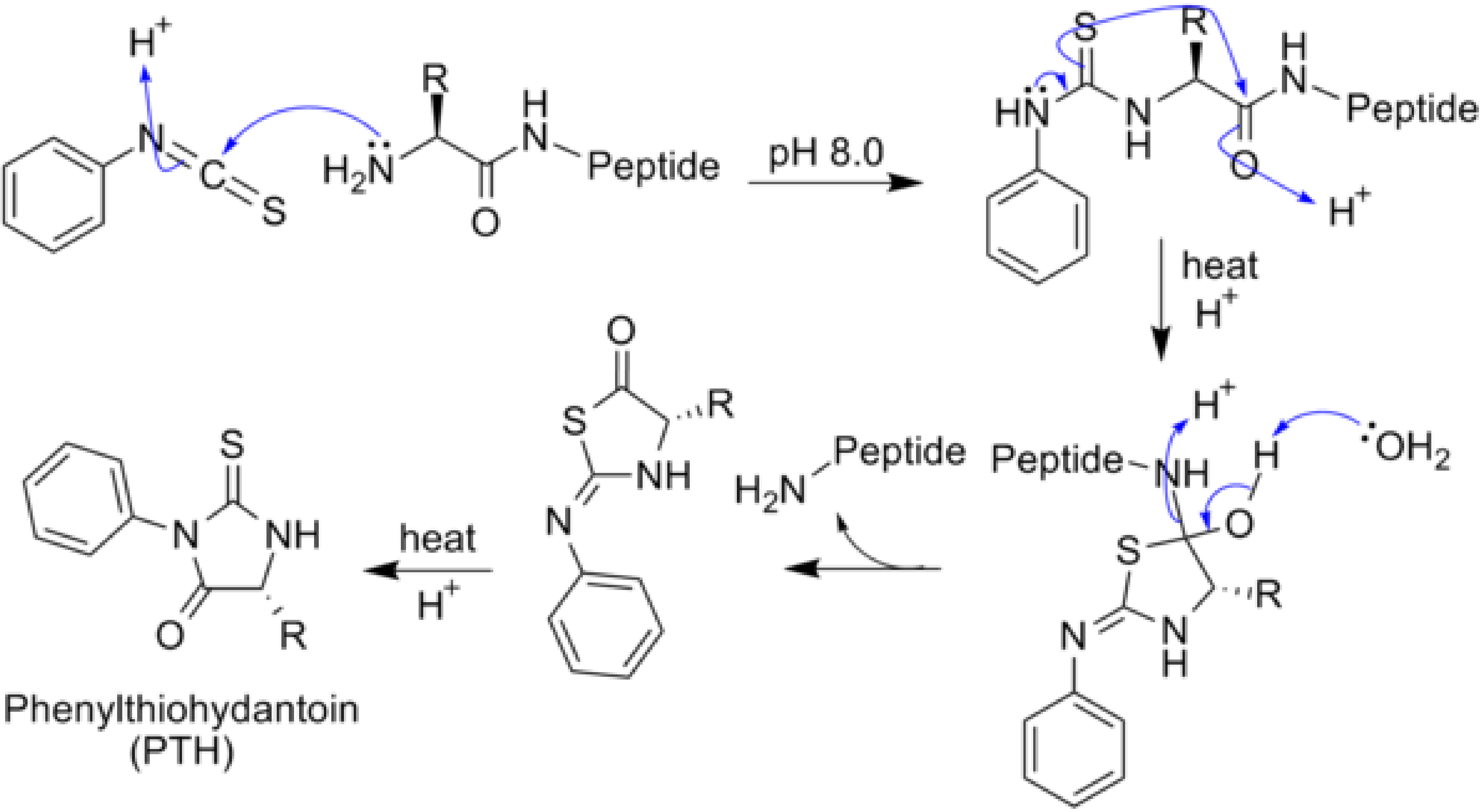
❑*Edman Degradation:-*

- Nucleophilic addition of the free amino group of the polypeptide to the C=N of phenyl isothiocyanate in a mild basic medium.

- Ring closure forms a N-phenylthiohydantoin which detaches itself from the rest of the peptide which remains intact with all its sequences.

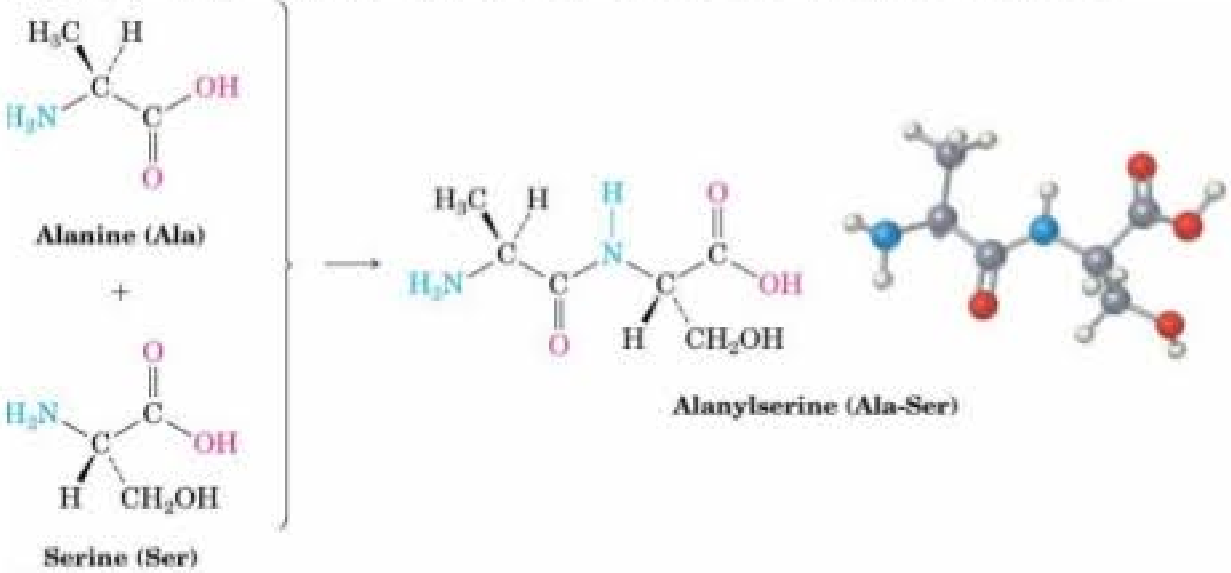
- N-phenylthiohydantoin is identified chromatographically by comparing with standards. This leads to identification of N-terminal amino acid.

- The residual peptide chain is subjected to Edman degradation repeatedly and all the amino acids in the polypeptide are identified.


H⁺
R
H
N
Peptide
pH 8.0
S
R
H
N
Peptide
H⁺
heat
H⁺
H⁺
Peptide
NH
H
OH₂
O
S
R
H₂N
Peptide
O
S
R
NH
S
N
NH
heat
H⁺
O
R
Phenylthiohydantoin
(PTH)

Peptides and Proteins

- Proteins and peptides are amino acid polymers in which the individual amino acid units, called residues, are linked together by amide bonds, or peptide bonds
- An amino group from one residue forms an amide bond with the carboxyl of a second residue



Alanine (Ala)

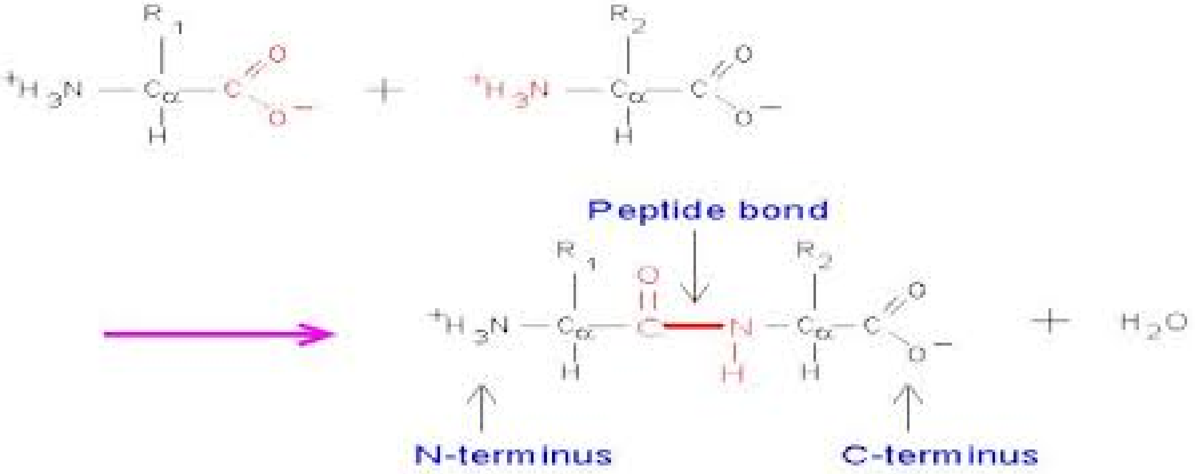
Serine (Ser)

Alanylserine (Ala-Ser)

Peptide Bonds

Are covalent bond between the α carboxyl group of one amino acid and the α amino group of another amino acid.

Peptide bonds are formed by the elimination of water.



I.3

N-Terminal and C-Terminal residue identification

Carboxypeptidase cleavage at the C-terminus