Department of Biochemistry and Clinical Biochemistry
State University of Medicine and Pharmacy "Nicolae Testemitanu"

AMINO ACIDS – STRUCTURE, CLASSIFICATION, PROPERTIES.

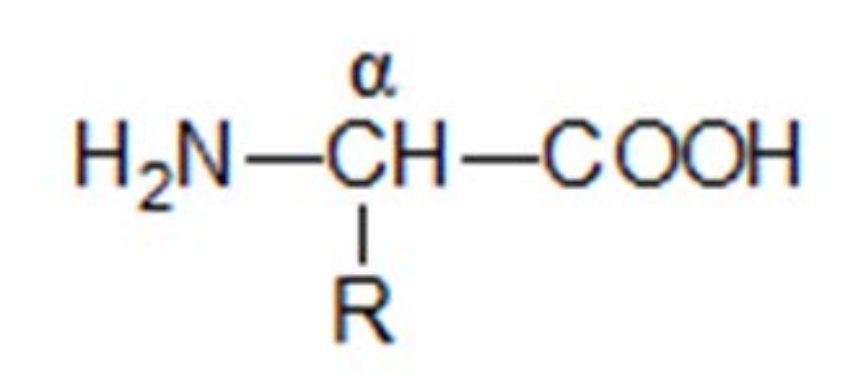
PRIMARY STRUCTURE OF PROTEINS

Amino acids are particularly important for the human body:

- are the basic structural elements of proteins;
- are precursors of the hormones, purine and pyrimidine nitrogenous bases, porphyrins, vitamins and biogenic amines.

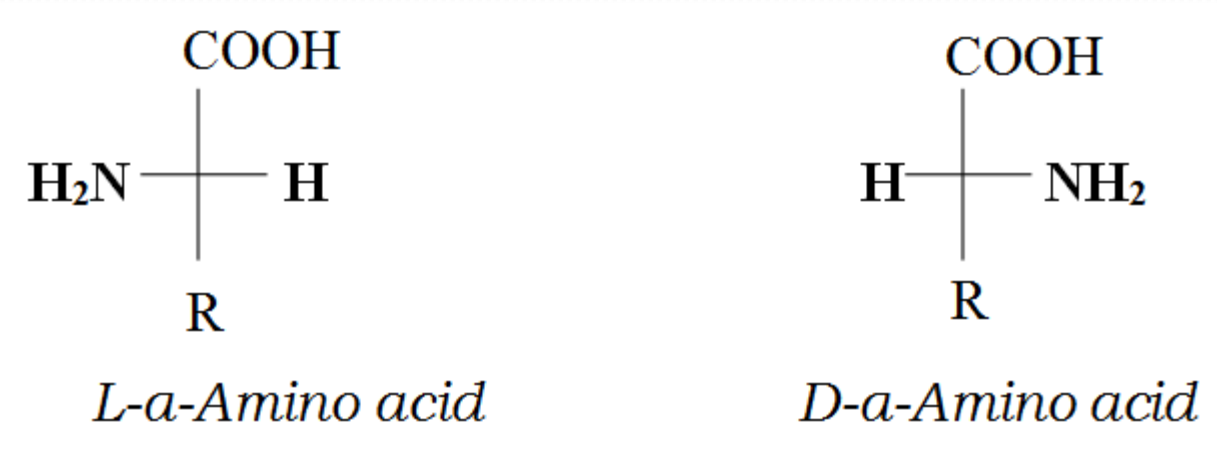
α-Amino acids are heterofunctional compounds containing carboxyl and amine groups linked to the same **α-**carbon atom. Amino acid radical (R) - is also joined to the **α-**carbon atom.

The general formula of α-amino acids is:



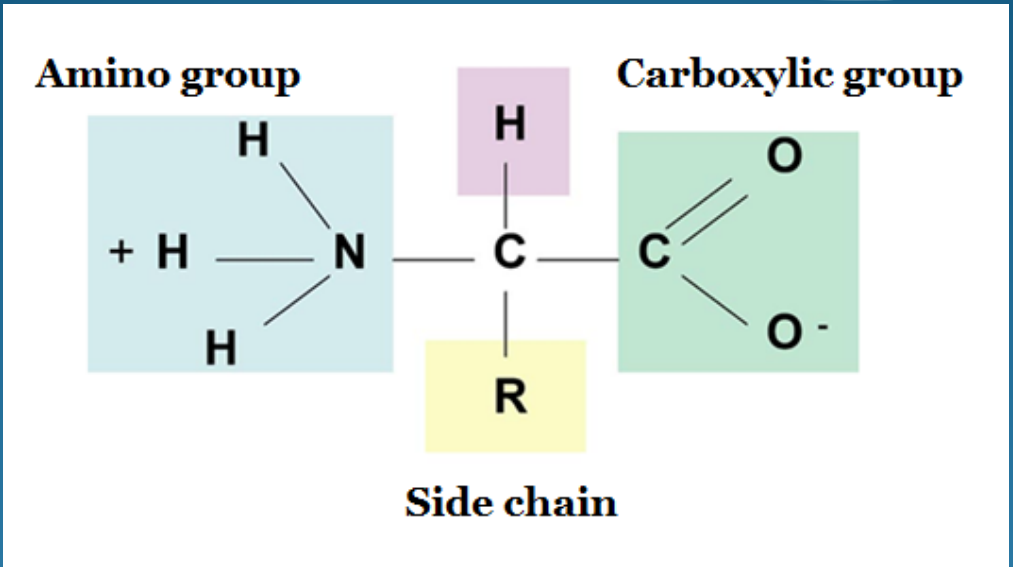
Stereoisomery of amino acids

- All representatives of α-amino acids (except glycine) contain a chiral α-carbon atom and form stereoisomers (enantiomers) - L and D.



- In the protein structure in living organisms only ***L-α-amino acids*** are used.

Acid-base properties of α-amino acids



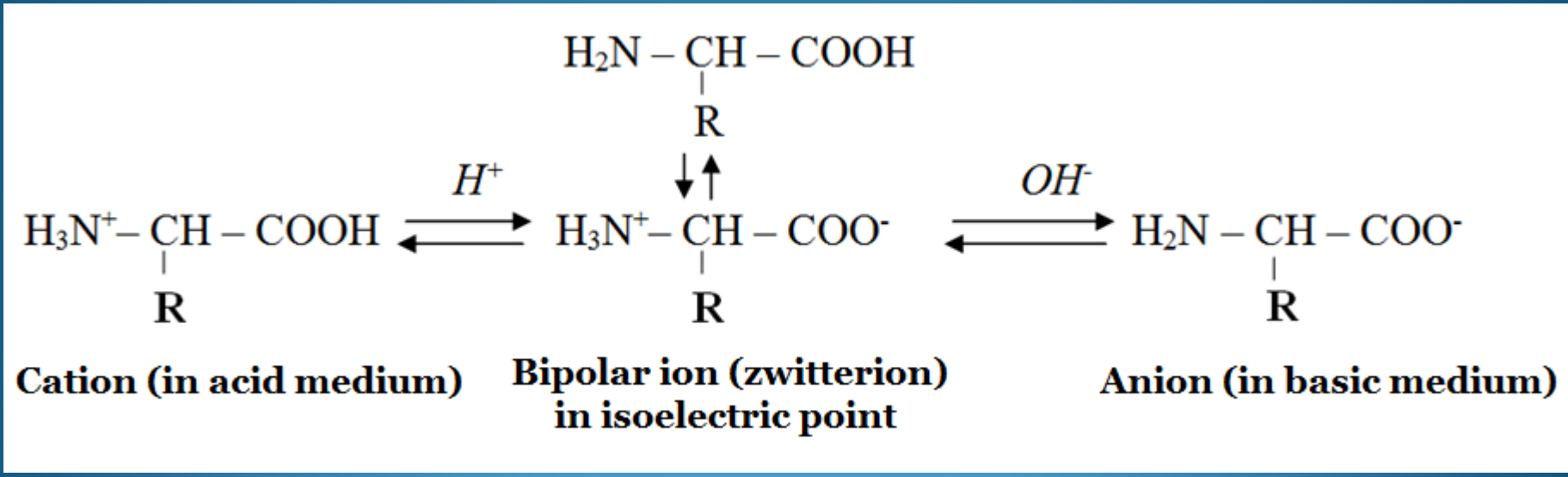
Under physiological pH, *in aqueous solution* α-amino acids exist in the form of **bipolar ions (zwitterions):**

- **the amino group is protonated** ($-NH_3^+$) and have basic properties - is a proton **acceptor;**
- **carboxyl group is dissociated (deprotonated)** ($-COO^-$) and has acidic properties - is a proton **donor**;

Thus, the amino acids have **amphoteric properties:** both - **basic and acidic**.

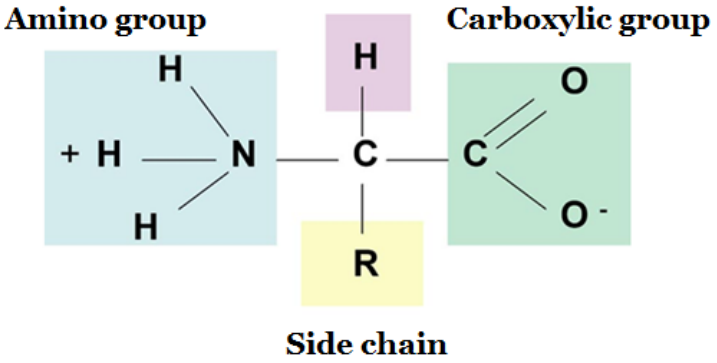
Acid-base properties of α-amino acids

In an acidic solution (pH <7) the amino acid is protonated and exist as a cation; in an alkaline solution (pH> 7), the amino acid is deprotonated, and exists as an anion. Thus, at some intermediate pH, the amino acid must be balanced exactly between the anionic and cationic forms and exists as a neutral bipolar ion (zwitterion). This pH is called **isoelectric point (Pi).**



In isoelectric point the zwitterion has a summary charge = 0 and is in an **isoelectric state**.

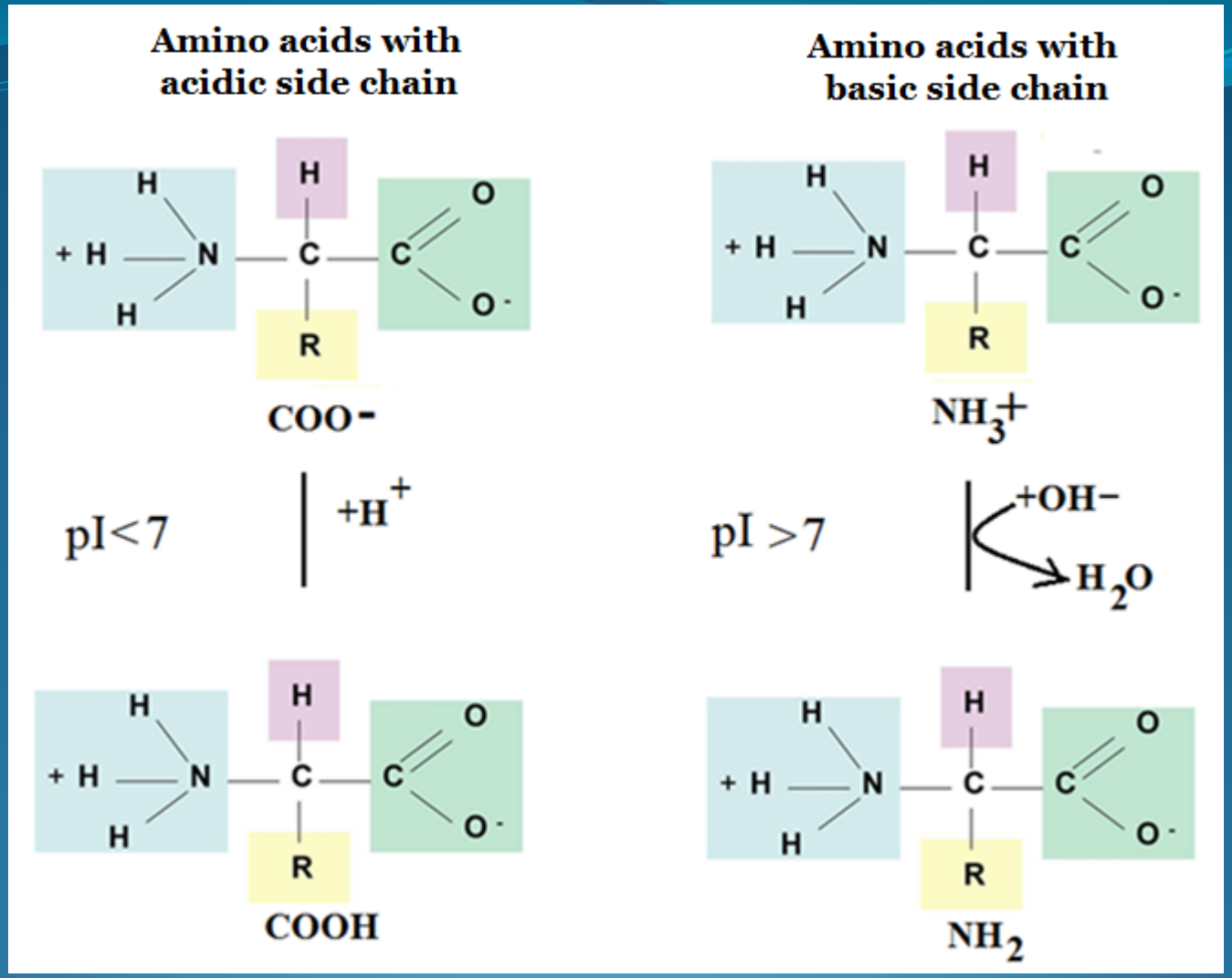
Acid-base properties of α-amino acids



Amino acids differ by **side chain (radical)**, which gives them specific properties. The radical can have ***hydrophobic or hydrophilic*** properties. Hydrophilic radicals may be ***neutral, acidic or basic***, depending on the functional groups.

In case the side chain of an amino acid is *hydrophilic or hydrophobic neutral* - it will not affect the total electric charge of the amino acid and its acidic or basic properties.

The hydrophilic acidic side chain in neutral medium have a *negative charge* and the amino acid is *an anion*. *The hydrophilic basic* side chain in neutral medium have a *positive charge* and the amino acid is *an cation*. In order to have such amino acids in the isoelectric state - it is necessary to change the pH of the medium.



Thus, the isoelectric point of the amino acids will vary from low values (pI <7) for acidic amino acids (pI=2.87 for aspartic acid) to high values (pI> 7) for basic amino acids (pI=10.8 for arginine). Neutral amino acids don't have the isoelectric point at a neutral pH, as it would be expected, but in week acidic medium (pI = 5-6).

Classification of amino acids according to the physico-chemical properties of the side chain

- 1. Amino acids with a **non-polar (hydrophobic) side chain**: glycine, alanine, valine, leucine, isoleucine, proline, phenylalanine, tryptophan and methionine. All of them are less soluble in water than the polar amino acids;

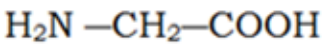
- 2. Amino acids with **a polar (hydrophilic) neutral side chain** (at pH = 6): serine, threonine, cysteine, tyrosine, asparagine, glutamine. These amino acids are more soluble in water than the non-polar amino acids, due to -OH, $-NH_2$, amide and -SH functional groups that can interact with water;

- 3. Amino acids with **a polar (hydrophilic) negatively charged side chain** (at pH = 6): aspartic acid and glutamic acid;

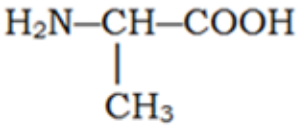
- 4. Amino acids with **a polar (hydrophilic) positively charged side chain** (at pH = 6): lysine, arginine, histidine.

Non-polar (hydrophobic) amino acids:

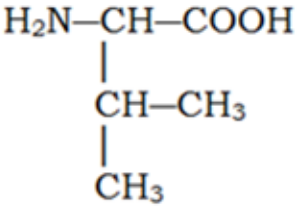
1. Glycine (Gly)



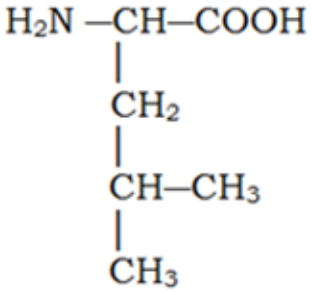
2. Alanine (Ala)



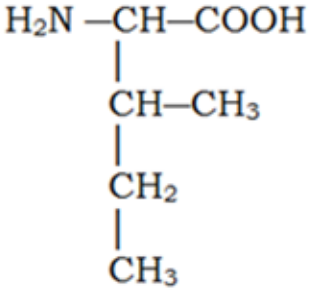
3. Valine (Val*)



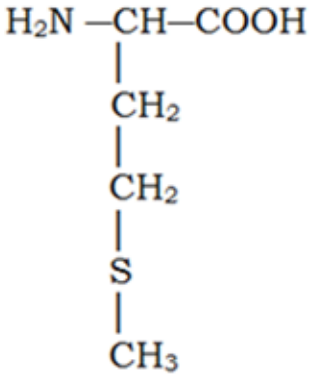
4. Leucine (Leu*)



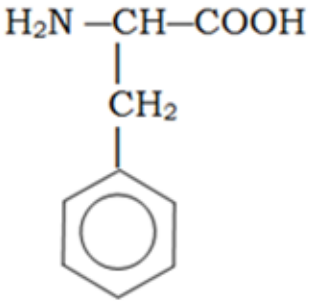
5. Isoleucine (Ile*)



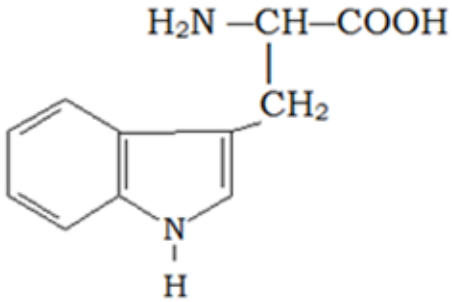
6. Methionine (Met*)



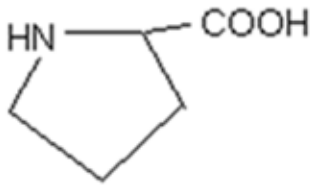
7. Phenylalanine (Phe*)



8. Tryptophan (Trp*)

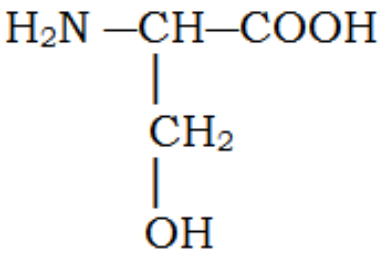


9. Proline (Pro)

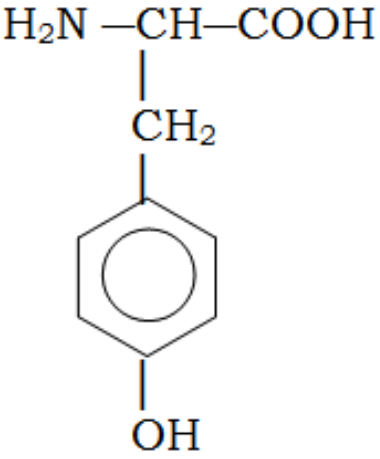


Polar (hydrophilic) neutral amino acids:

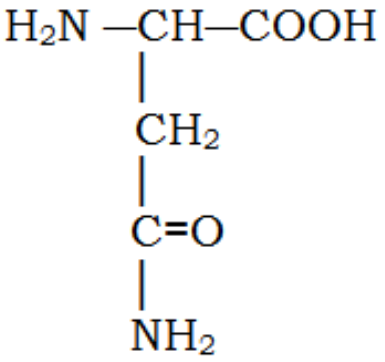
10. Serine (Ser)



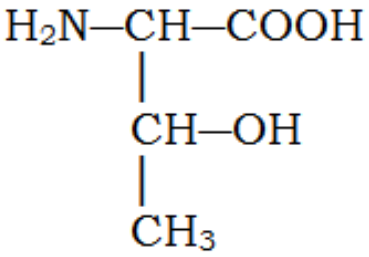
12.Tyrosine (Tyr)



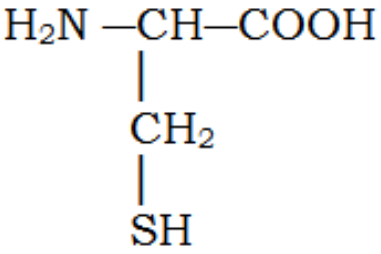
14. Asparagine (Asn)



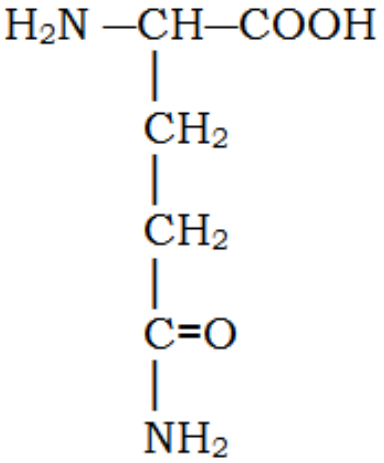
11. Threonine (Thr*)



13. Cysteine (Cys)

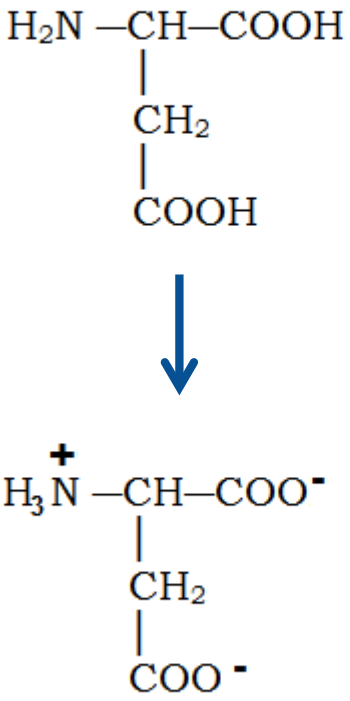


15. Glutamine (Gln)

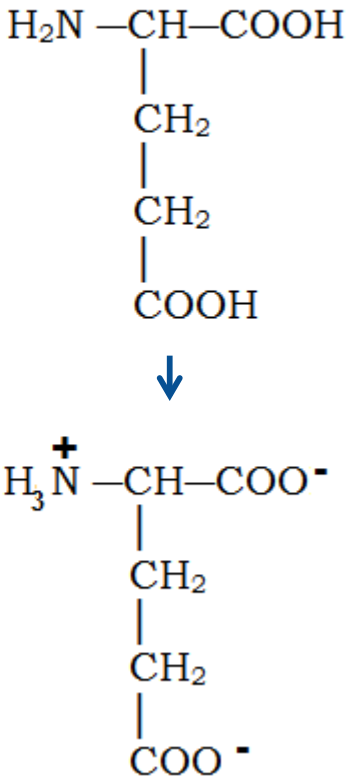


Polar (hydrophilic) negatively charged (acidic) amino acids:

16. Aspartic acid (Asp)

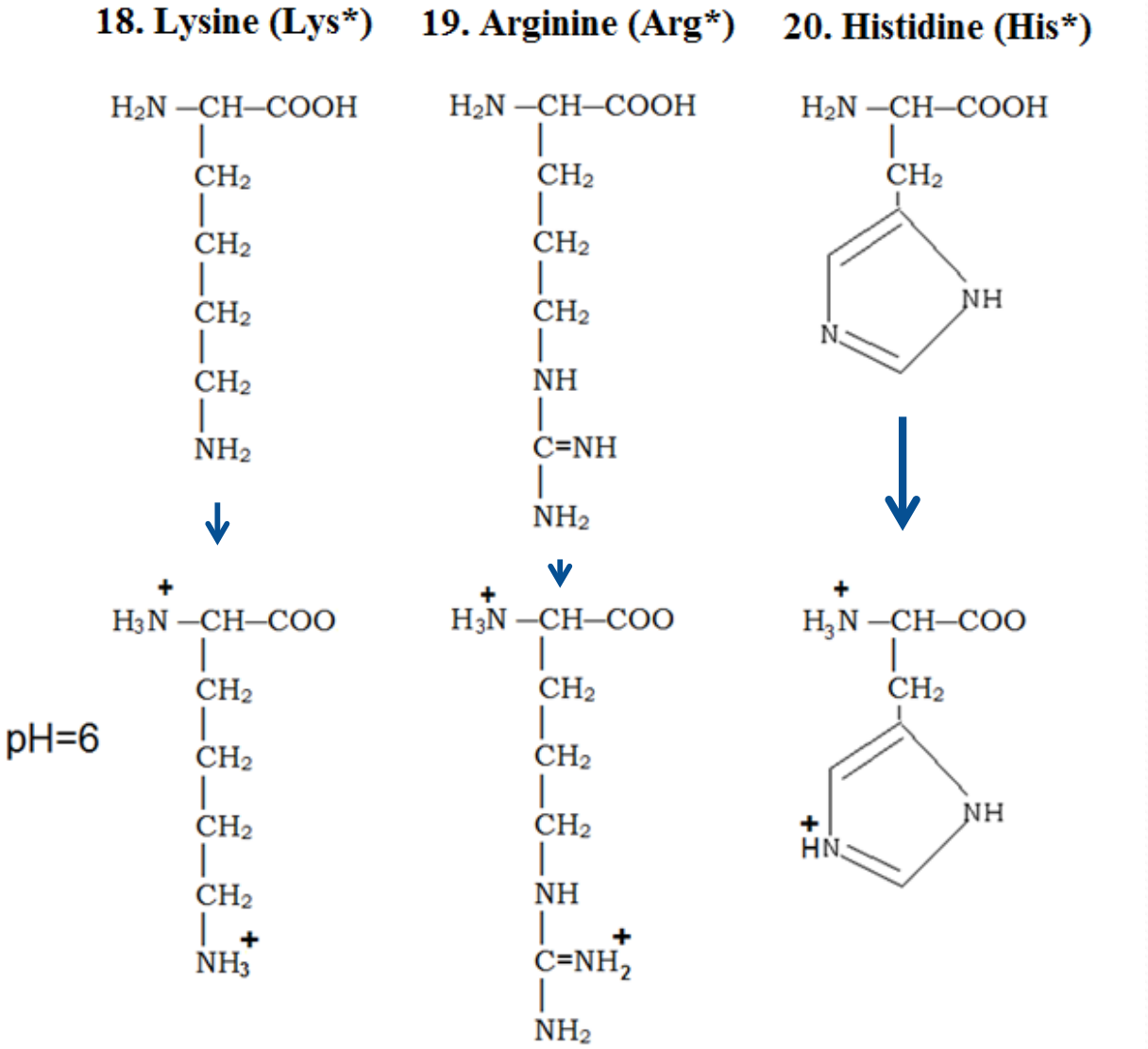


17. Glutamic acid (Glu)



pH=6

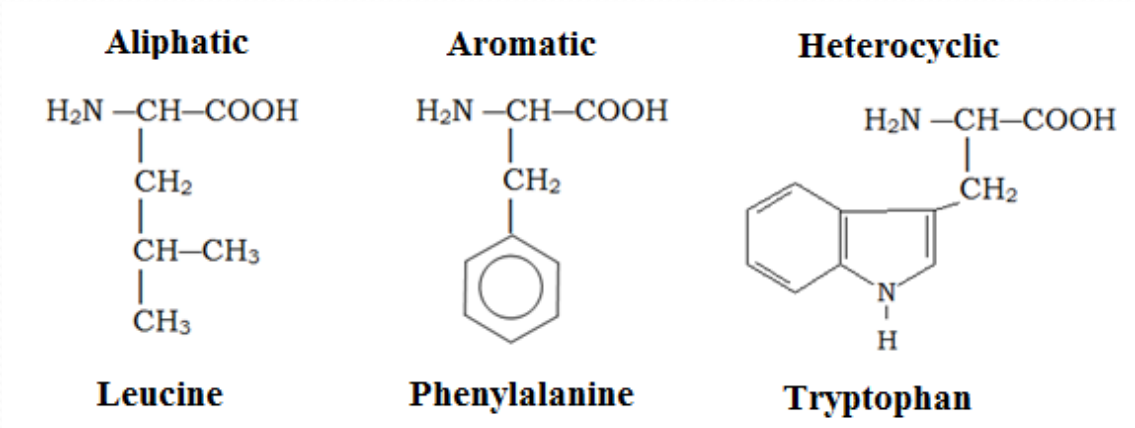
Polar (hydrophilic) positively charged (basic) amino acids:



Classification by chemical structure:

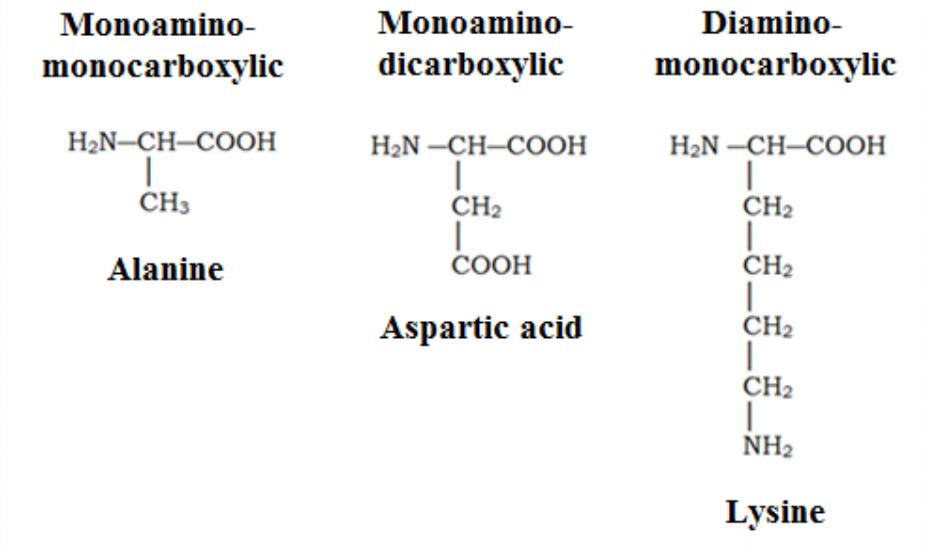
- by the chemical structure of the side chain:

Examples:



- By number of groups –COOH and –NH2:

Examples:

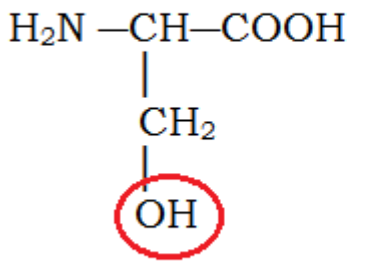


Classification by chemical structure:

- by the presence of other functional groups in the chain –

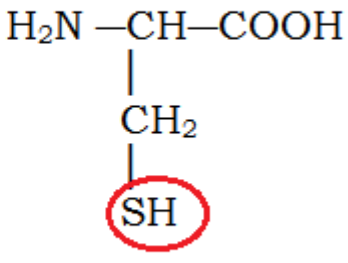
Examples:

Hydroxy amino acids



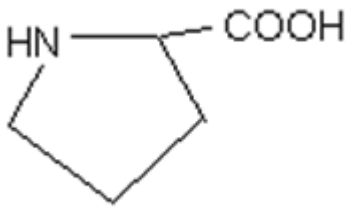
Serine

Thio amino acids



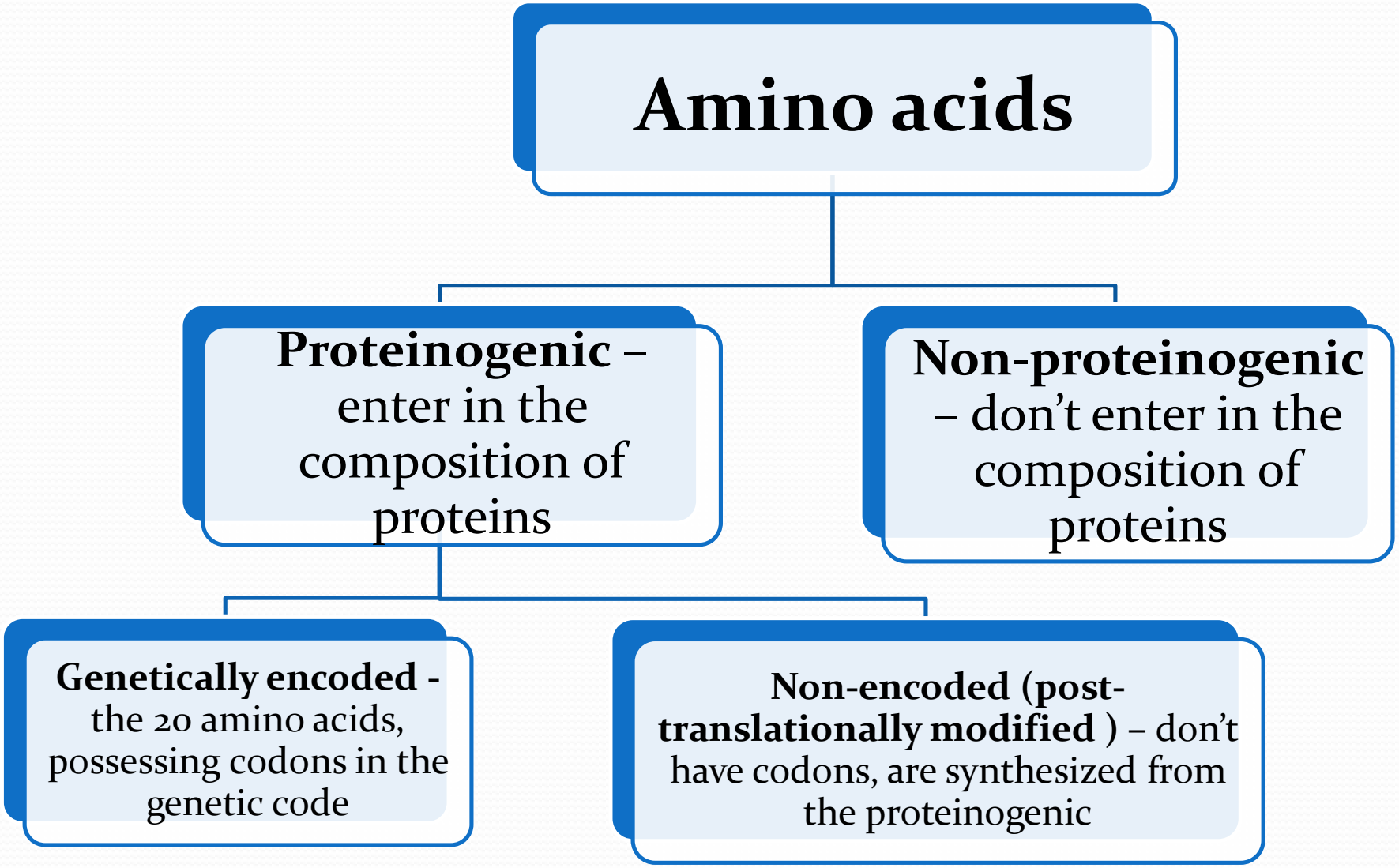
Cysteine

- Imino acid -

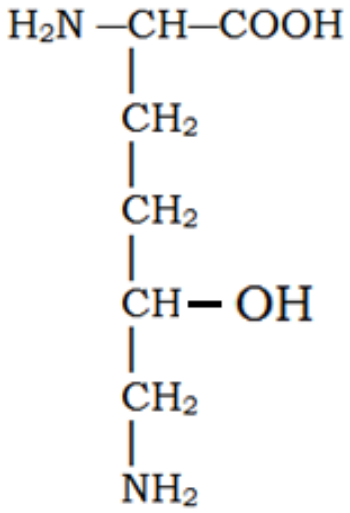


Proline

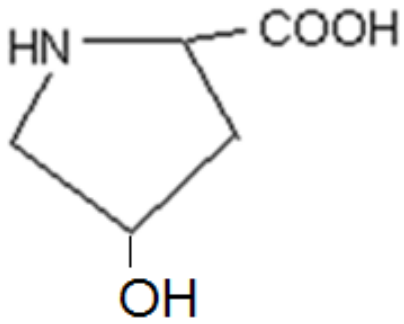
Classification of amino acids by the presence in protein and genetic encoding



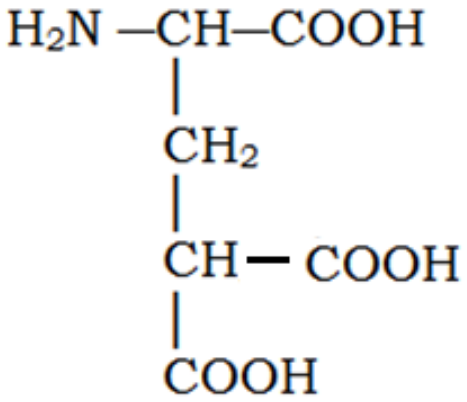
Proteinogenic non-encoded (post-translationally modified) amino acids:



Hydroxylysine

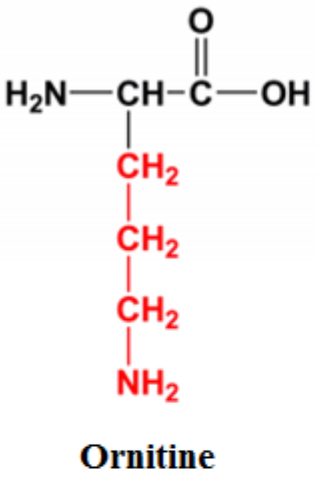


Hydroxyproline

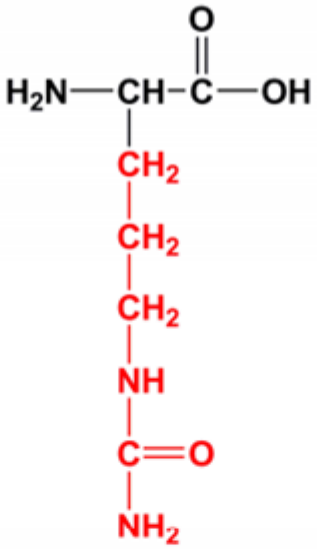


γ-carboxyglutamic acid

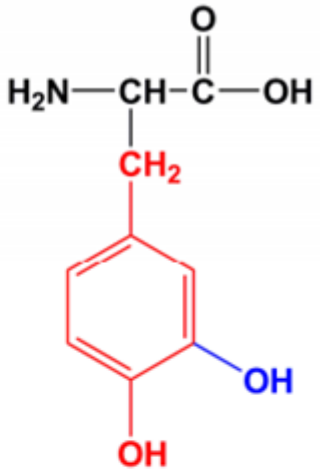
Non-proteinogenic amino acids – don't enter in the composition of proteins:



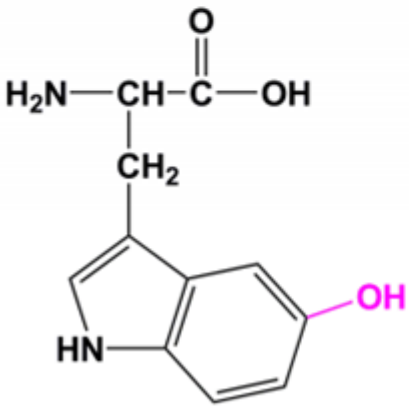
Ornitine



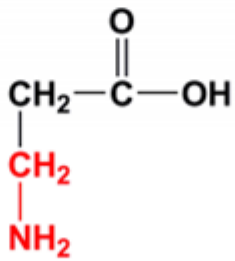
Citrulline



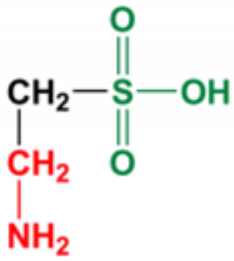
Dopa



5-hydroxy tryptophan



β-alanine



Taurine

Classification by biological role:

Amino acids

Essential:

Valine*
Leucine*
Isoleucine*
Phenylalanine*
Triptophan*
Methionine*
Threonine*
Lysine*

They are not synthesized in the body, it is necessary to receive them with food

Semi-essential:

Arginine*
Histidine*

They are synthesized in the body, but in insufficient quantities, it is necessary to receive them with food

Non-essential:

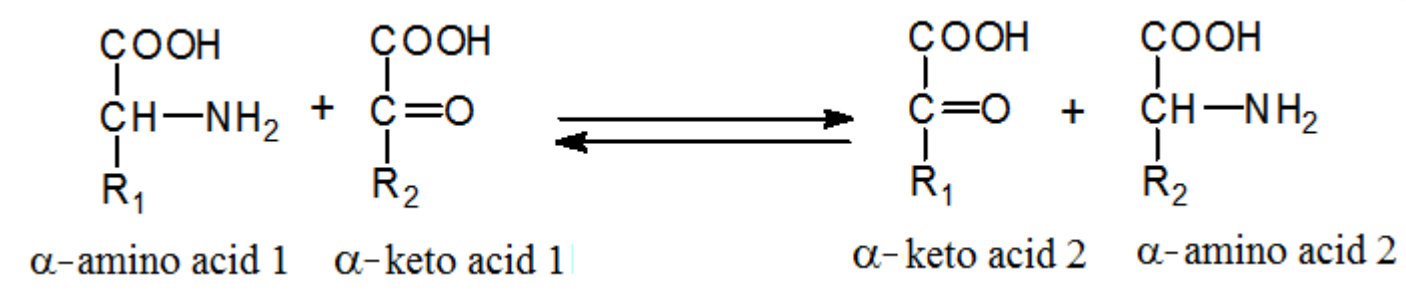
Glycine
Alanine
Proline
Serine
Tyrosine
Cysteine
Asparagine
Glutamine
Aspartic acid
Glutamic acid

They are synthesized in the body

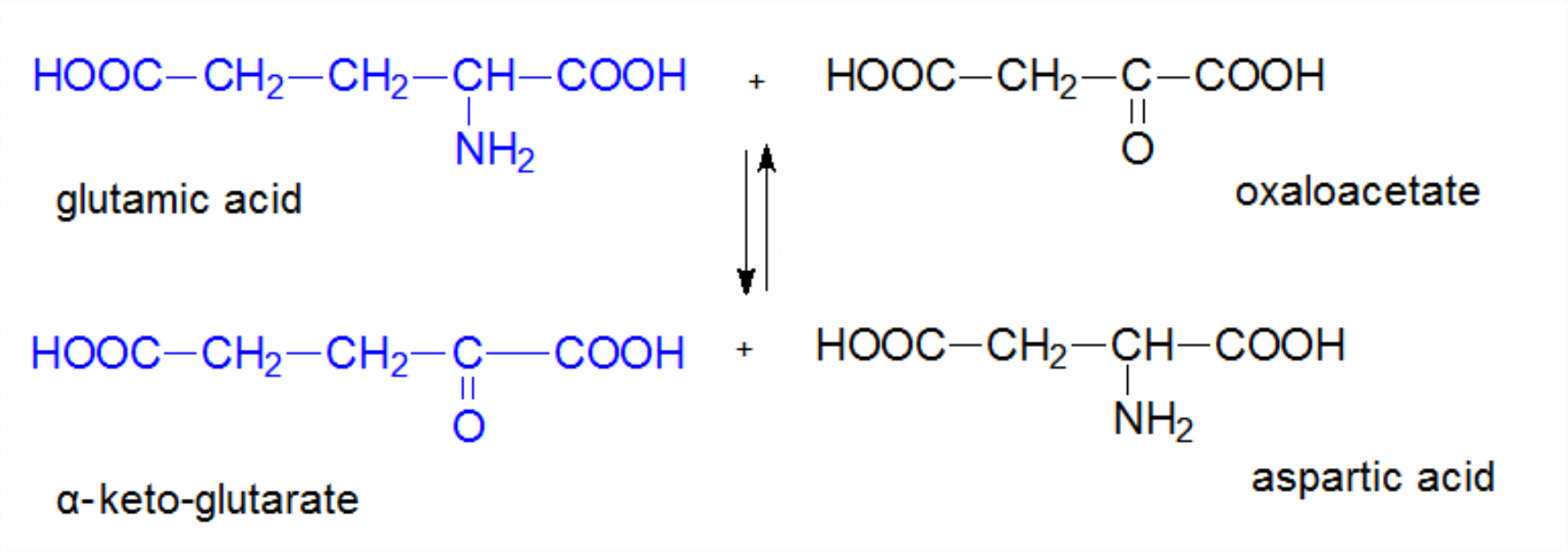
Reactions of biological importance

1. ***Transamination of α-amino acids:***

- is the transferring of the amino group from the *donor* amino acid to an α-keto acid – *acceptor* of the amino group. In this reaction the α-amino acid is converted to a keto acid, and the keto acid is converted to an amino acid:



For example, the transamination reaction between glutamic acid and oxaloacetate: it produces a new amino acid - aspartic acid and a new keto acid - α-ketoglutarate:



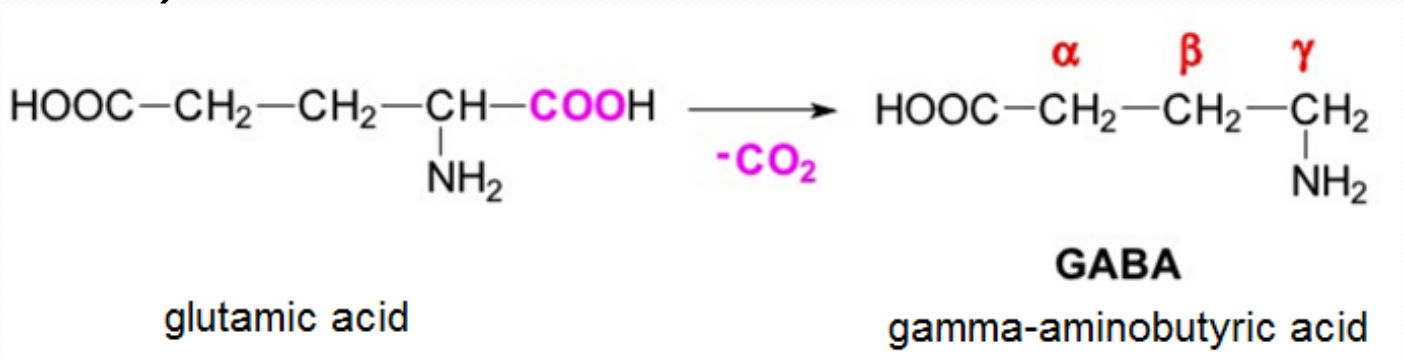
Reactions of biological importance

2. *Decarboxylation of α-amino acids:*

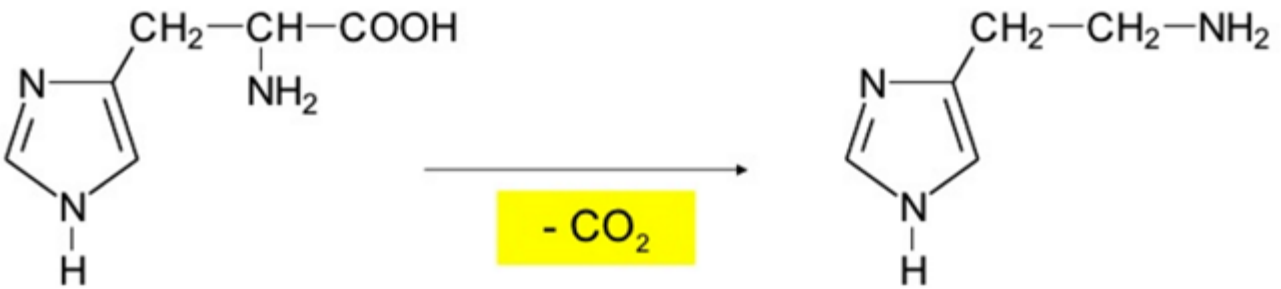
- in the decarboxylation reaction the α-amino acids lose the α-carboxylic group and are converted into biogenic amines

For example:

a) decarboxylation of glutamic acid and gamma-aminobutyric acid (GABA) formation:



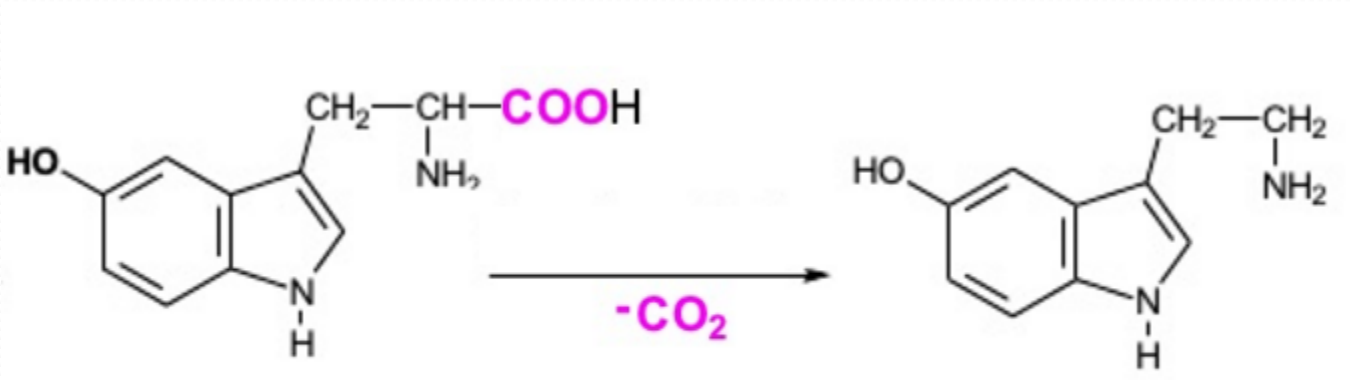
b) Decarboxylation of histidine and histamine formation:



Histidine

Histamine

C) Decarboxylation of 5-hydroxy tryptophan and serotonine formation:

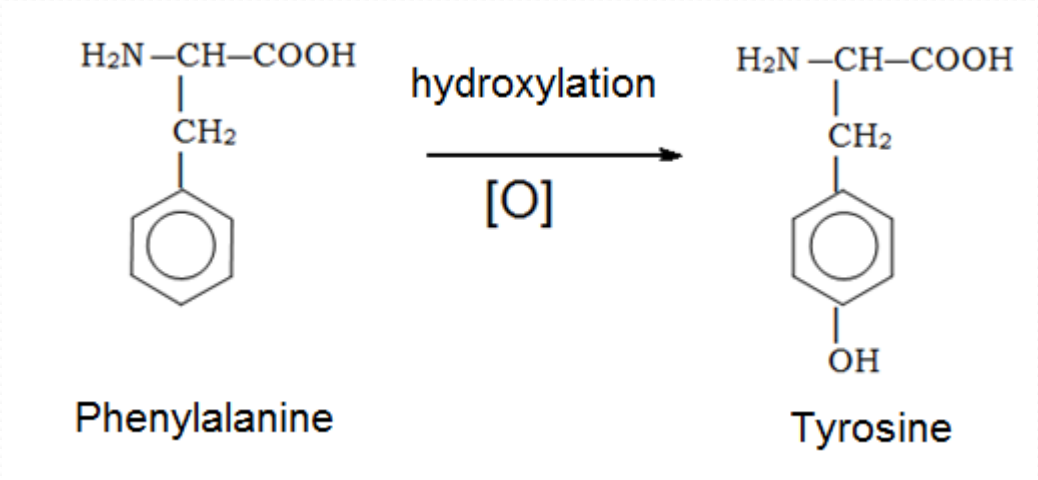


5-hydroxy tryptophan

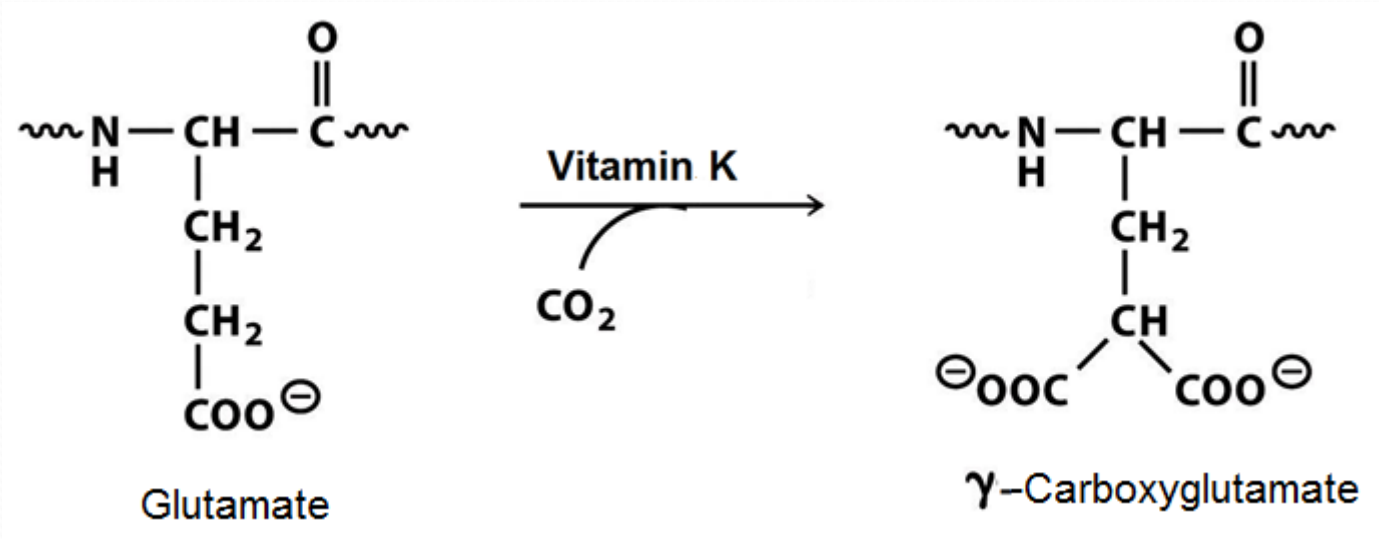
Serotonine

Reactions of biological importance

3. *Hydroxylation of α-amino acids:*

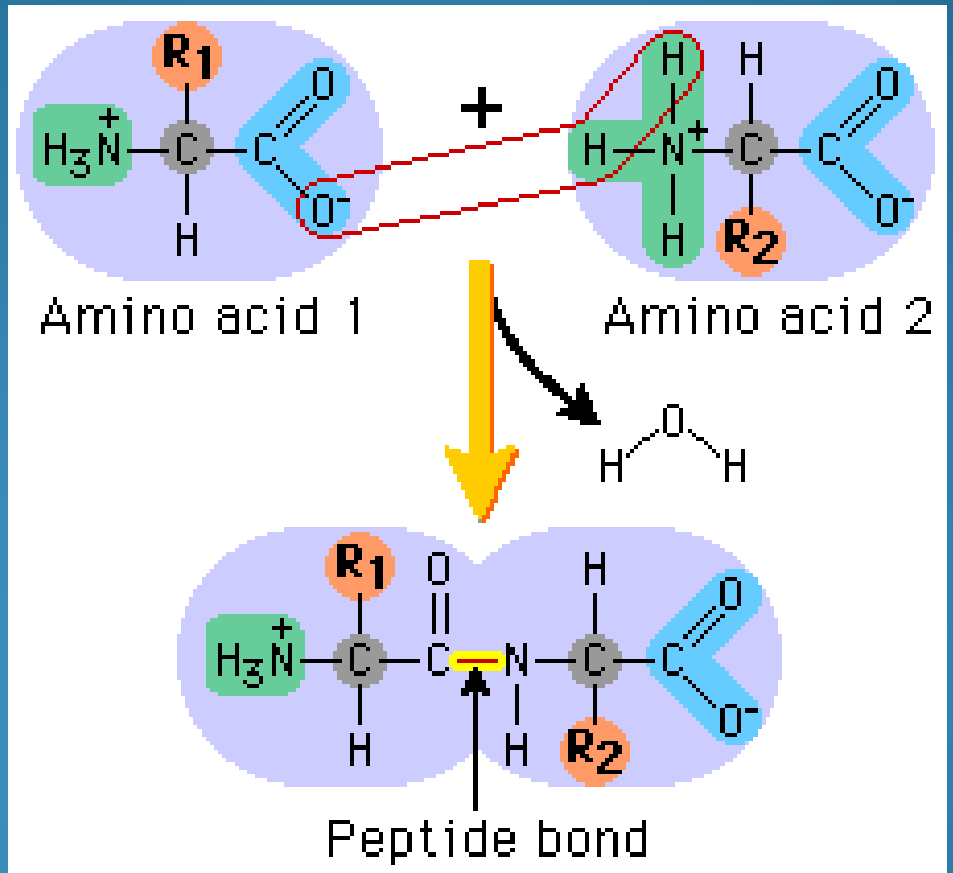


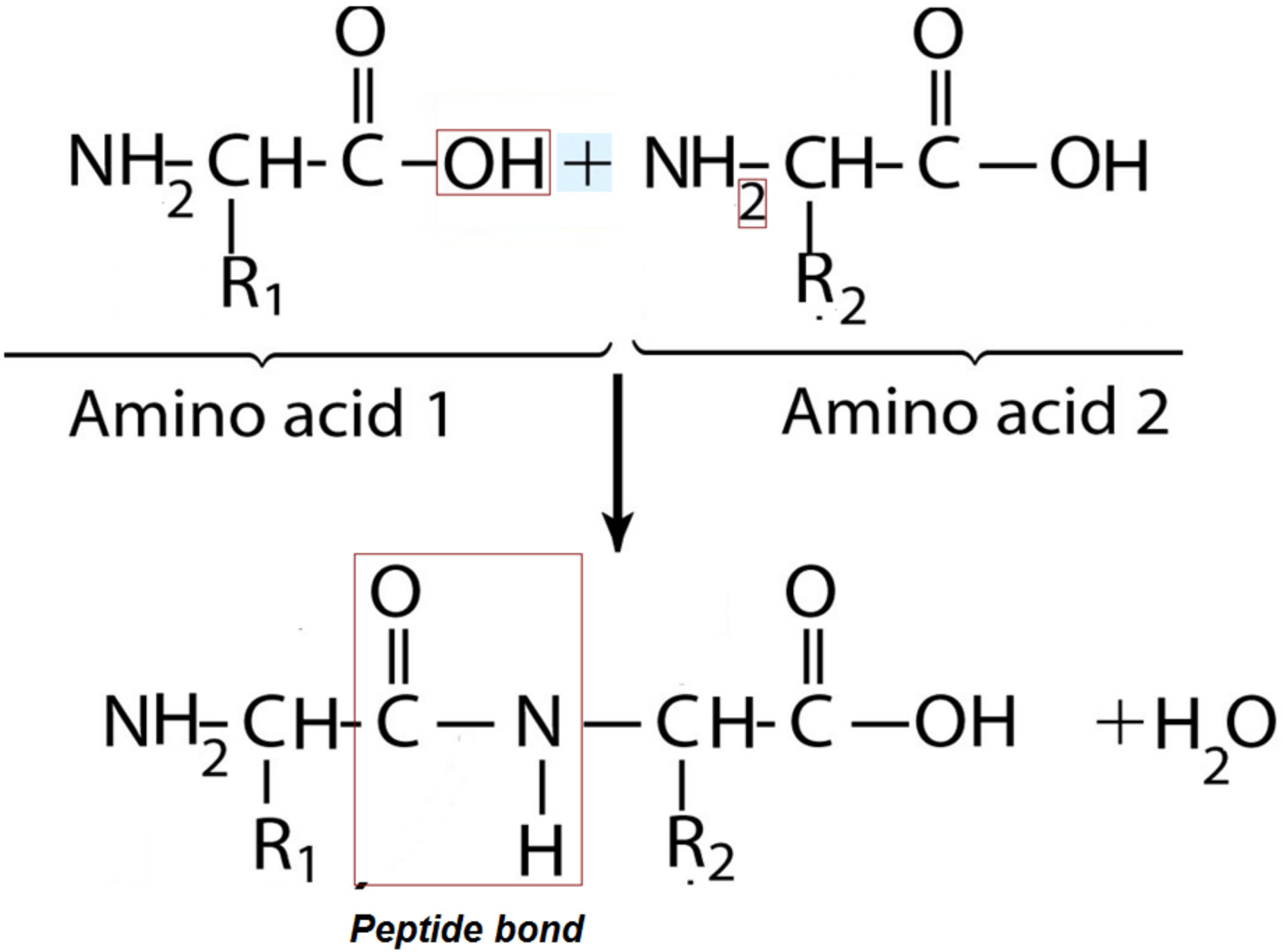
4. *Carboxilation of amino acids:*



The amino acids are joined in the chains by peptide bonds

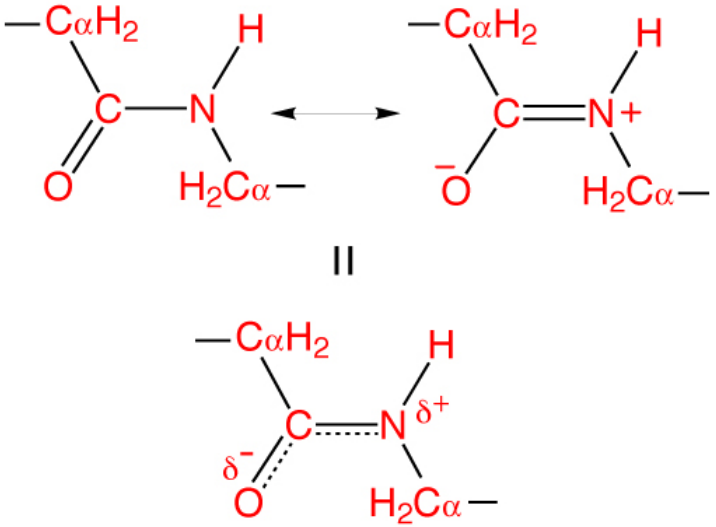
The peptide bond is formed between the α-carboxylic group of one amino acid and the α-amino group of the following amino acid :



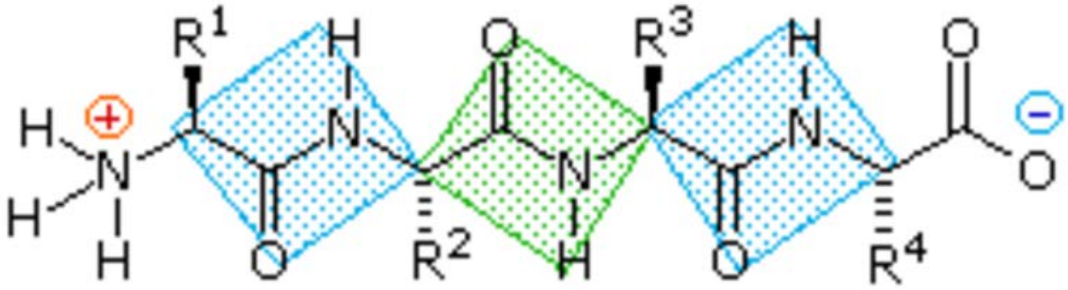

Amino acid 1
Amino acid 2
Peptide bond

The properties of peptide bond:

- The classical peptide bond is a **strong covalent bond** and has the properties of partially double bond.

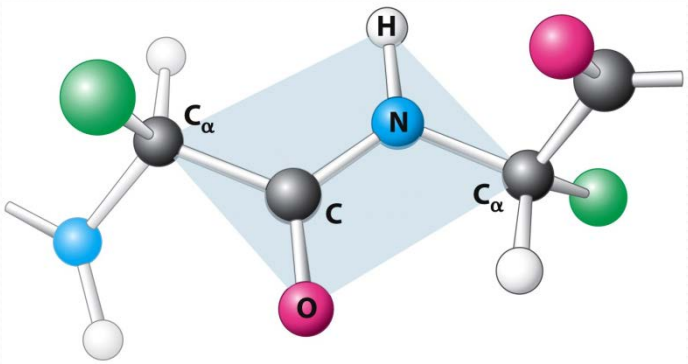


- The peptide bond is **planar** - all the atoms of the peptide group are in the same plane.

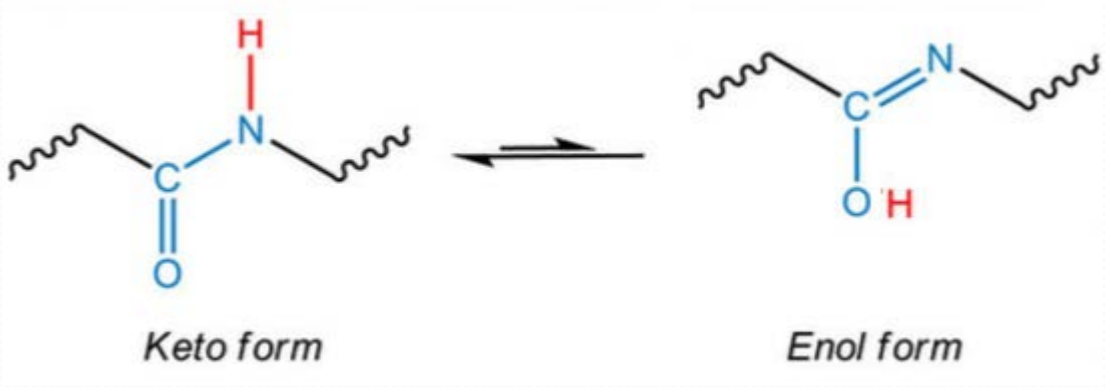


The properties of peptide bond:

- The classical peptide bond has **trans- conformation**.

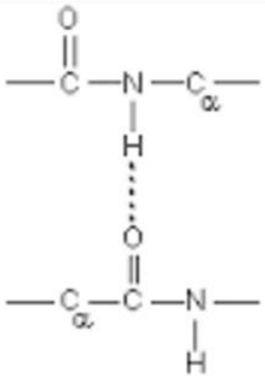


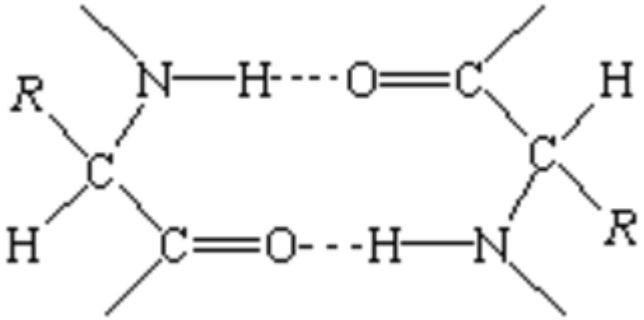
- Peptide bond has two resonance forms - **keto and enol** :



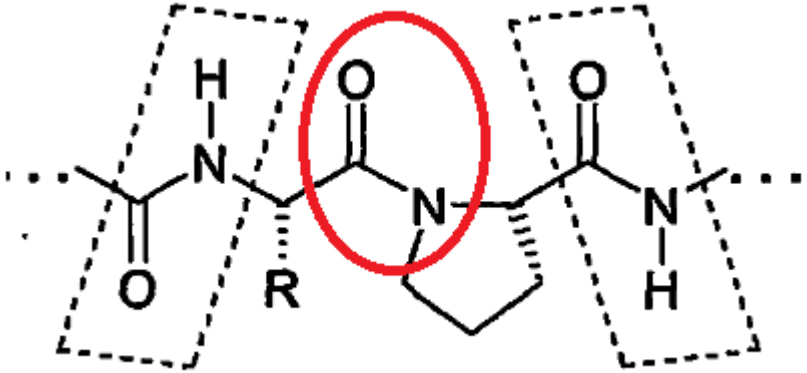
The properties of peptide bond:

- Each classical peptide bond is capable of forming **2 hydrogen bonds** with other polar atoms.



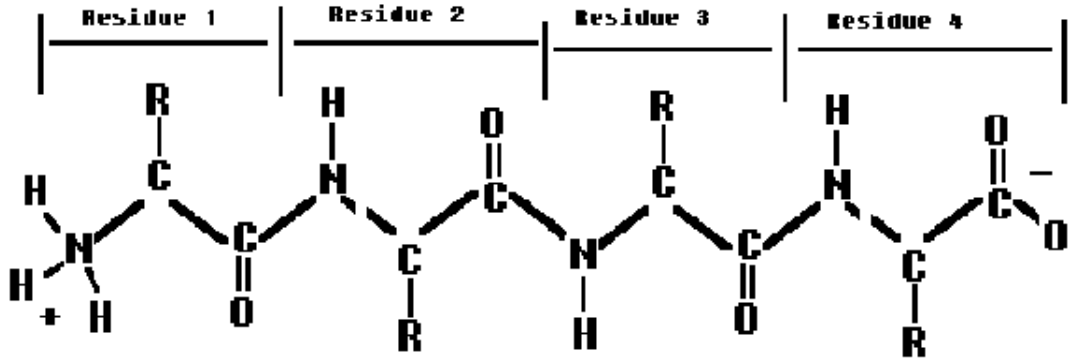


- Proline forms an **atypical peptide bond :**



- The products of the polycondensation of -amino acids linked by peptide bonds are called **peptides**:
- A peptide containing two amino acids is called a **dipeptide**; containing three amino acids – a **tripeptide**; and so on.
- A chain containing up to 50 amino acids, is called **oligopeptide**; containing 50-100 amino acids - **polypeptide**;
- If the number is greater than 100 amino acids, the polypeptide is called **protein**.

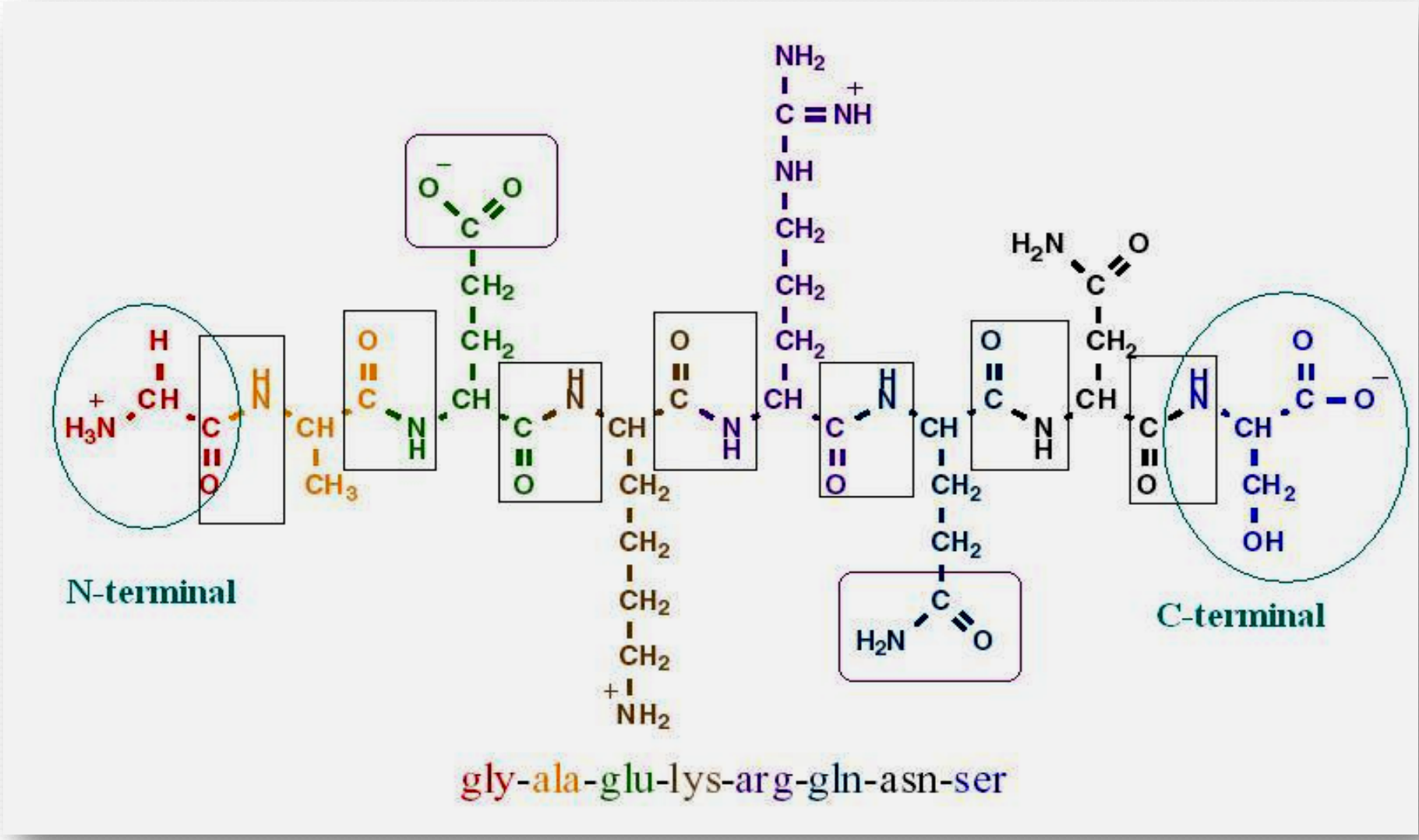
- The end of the peptide chain with the **-NH2 group** is known as the **N-terminal**, and is considered the beginning of the chain; and the end with the **-COOH group** is the **C-terminal**.
- The conformation of a peptide chain has a form of a zig-zag :



- The "R" groups come from the 20 amino acids which occur in proteins. The peptide chain is known as the ***backbone***, and the "R" groups are known as ***side chains***.
- "R" – side chains of the amino acids - they are maximal distant in space from each other.
- Each protein has a specific sequence of amino acids which are assembled under the direction and control of nucleic acids.

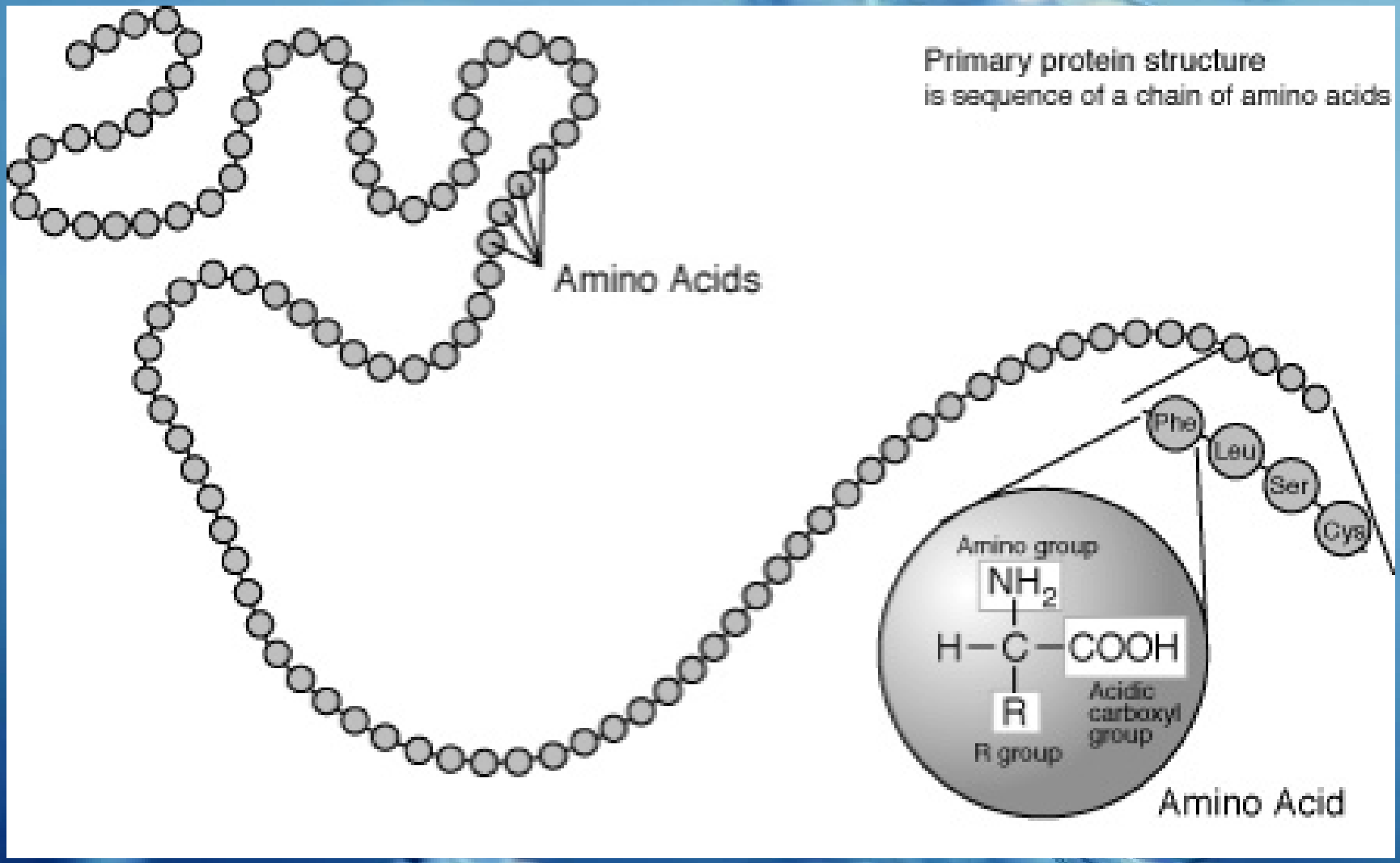
Nomenclature of peptides

All amino acids in the polypeptide chain situated on the left to the C-terminus have **–yl** terminus and the C-terminal keeps its trivial name. For example: tripeptide Gly-Ala-…-Ser will be called **glycyl-alanyl-…-serine.**



The sequence of amino acids in a protein is called

primary structure of protein

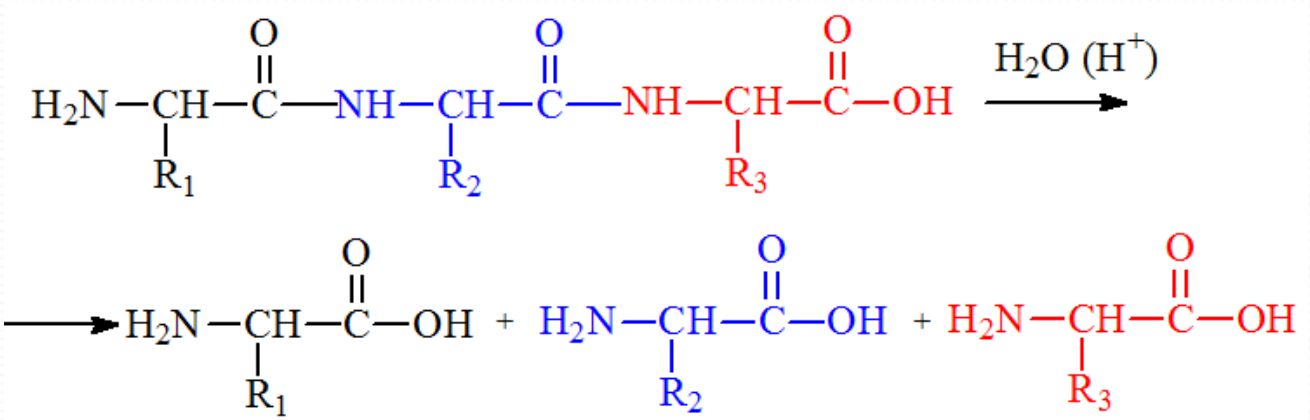


Determination of the primary structure of proteins

It has two main steps:

1. Determination of the amino acid **composition** of the peptide or protein;
2. Determination of the **sequence** of amino acids into polypeptide chain.

•The amino acid composition is determined by the analysis of **protein hydrolysates**. The total hydrolysis may be carried out by boiling the protein in a solution of 6M hydrochloric acid or by enzymes. All the peptide bonds are cleaved. For example:



•The determination of each amino acid in the hydrolyzate is carried out by **chromatography**. Currently such an analysis is performed automatically using special devices called **amino acid analyzers**.

Determination of the primary structure of proteins

The sequence of amino acid is determined in several stages:

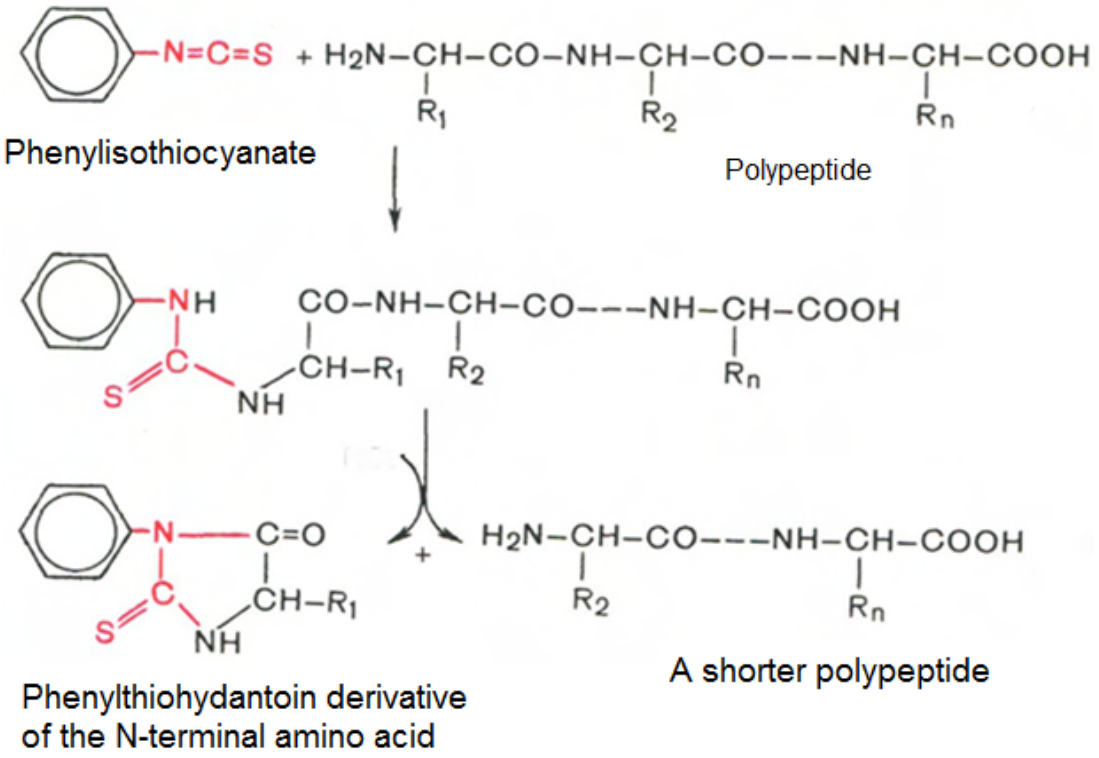
1. - selective partial hydrolysis of the polypeptide in shorter peptides (via several enzymatic or chemical methods);

2 - sequential identification of α-amino acids from the N- or C-terminal end for each peptide; as a rule the Edman method is applied;

3. – determination of the peptide order in polypeptide by overlapping and determination of the coincidence segments ("fingerprints“ method or "peptide mapping method“

Edman Method

- consists in the interaction of N-terminal amino acid with **phenylisothiocyanate** in weak basic medium. At a subsequent treatment with a weak acid without heating a cleavage of the N-terminal amino acid as a **phenylthiohydantoin derivative** occurs. This compound can be further identified by chromatographic method. This procedure is repeated several times until complete cleavage of the peptide fragment takes place:



Edman method has been shown to be useful for reproduction in an automatic device called **sequencer.** It can performe 40-50 cleavage steps.