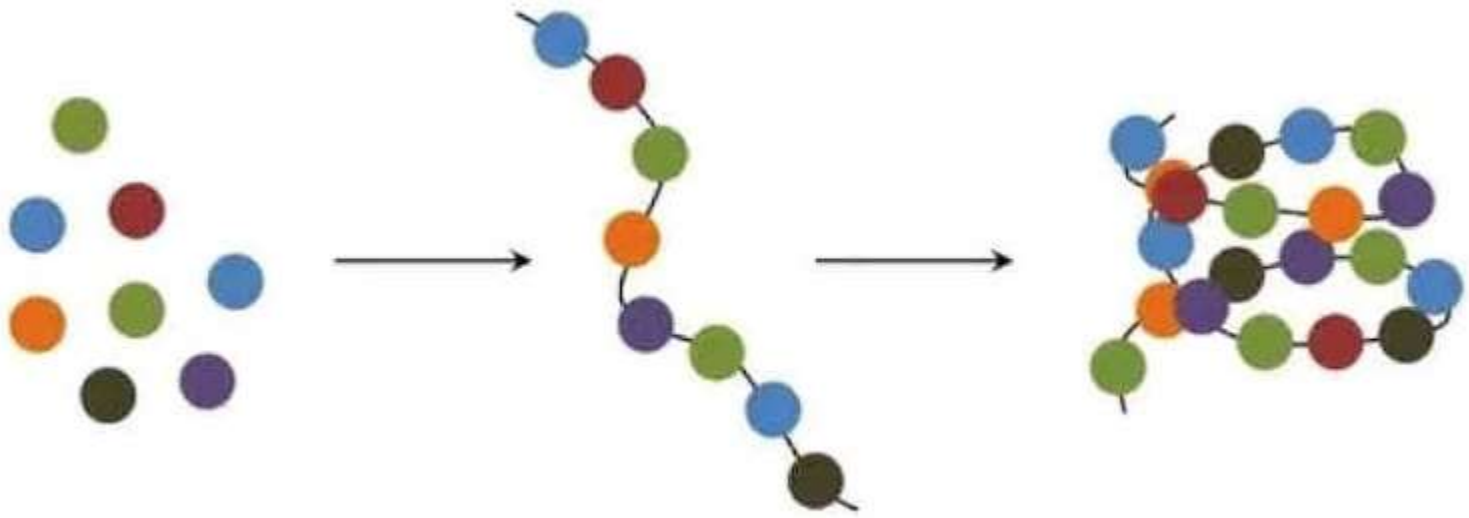
«Amino acids, peptides, proteins»



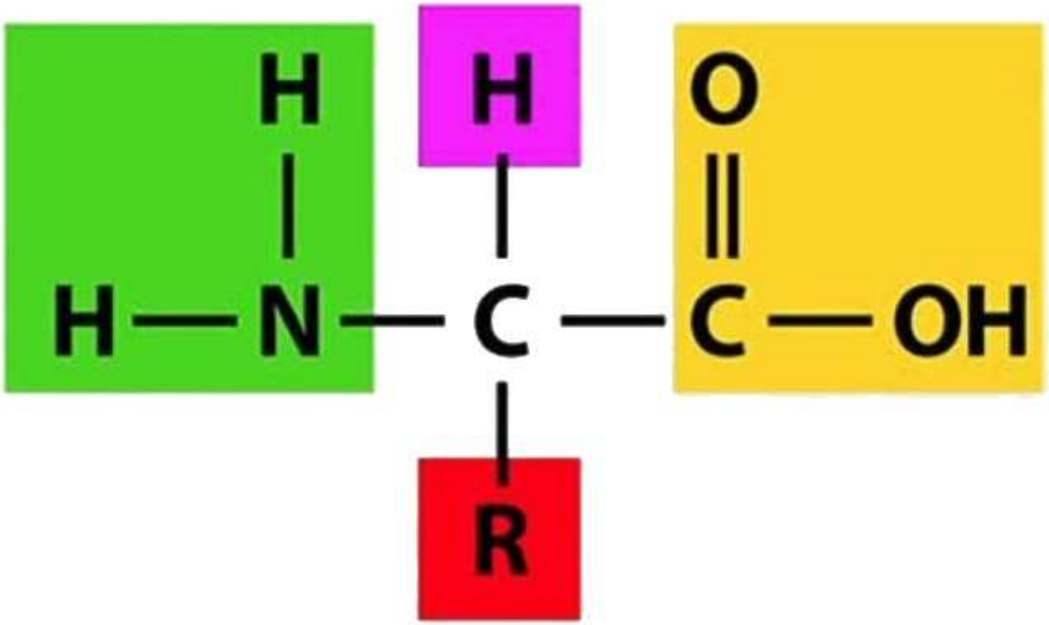
Ministry of Public Health of Ukraine
Poltava State Medical University

Lecture plan

1) Structure and classification of amino acids
2) Physico-chemical properties of proteinogenic amino acids
3) Natural peptides: classification, biochemical characteristics
4) Structure and classification of proteins
5) Physico-chemical properties of proteins
6) Levels of structural organization of protein molecules
7) Biological functions of proteins and peptides

AMINO ACIDS are heterofunctional compounds that contain two functional groups: a carboxyl group - **COOH** and an amino group - NH_2, associated with a hydrocarbon radical

General Formula



Hydrolysis of natural proteins and peptides releases about 20 different α-L-amino acids, the location of each of which in the polypeptide chain is encoded by a triplet of nucleotides in the DNA of the genome.

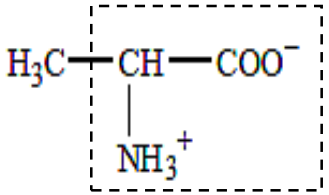
AMINO ACIDS ARE DIVIDED INTO

Proteinogenic (20 amino acids that are part of natural proteins and peptides)

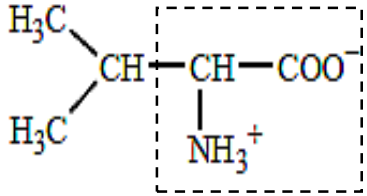
Non-proteinogenic or natural (there are more than 150, are not part of natural proteins and peptides).

Proteinogenic amino-acids

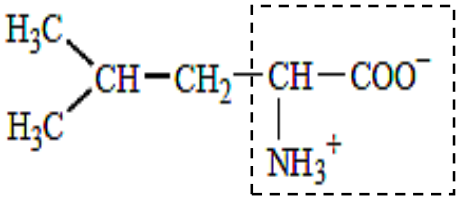
The frame is surrounded by a part that is the same for all amino acids. Amino acids differ in the structure of the side chain (R-group).



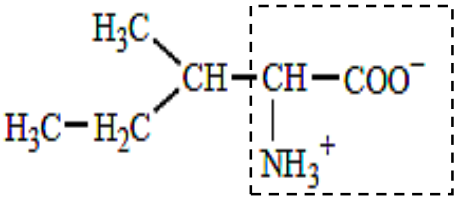
Alanine



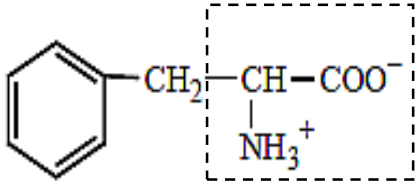
Valine



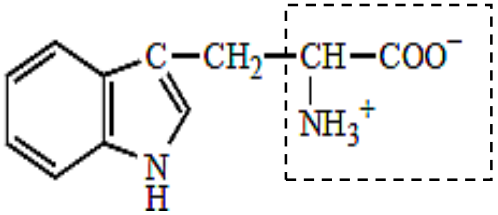
Leucine



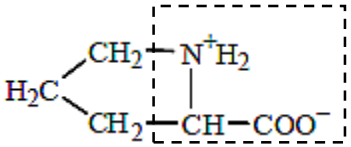
Isoleucine



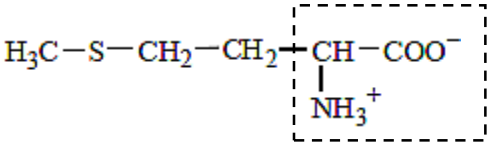
Phenylalanine



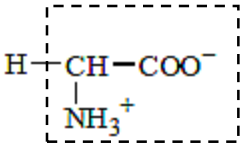
Tryptophan



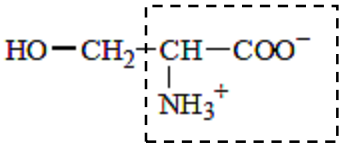
Proline



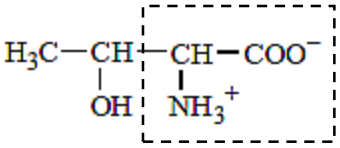
Methionine



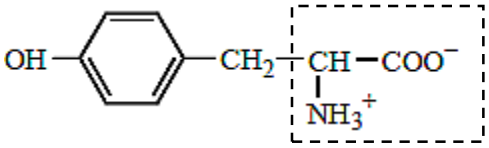
Glycine



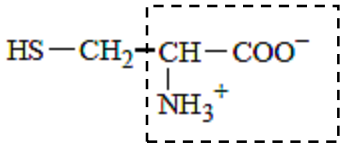
Serine



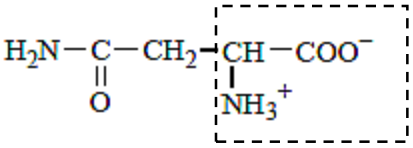
Threonine



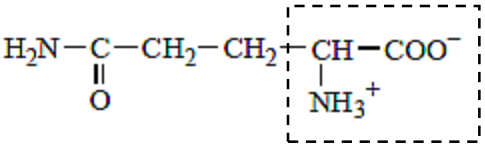
Tyrosine



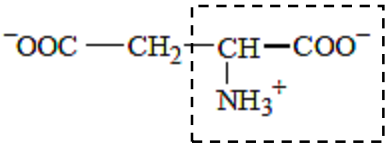
Cysteine



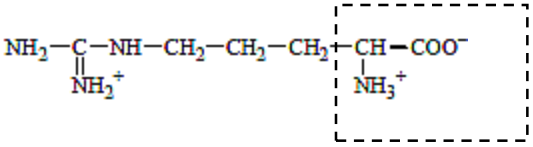
Aspargine



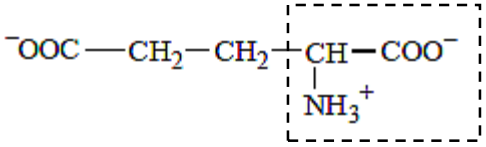
Glutamine



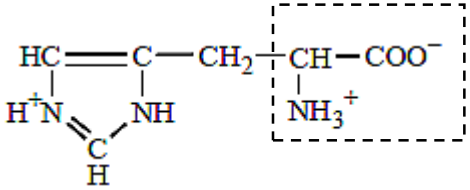
Aspartic acid



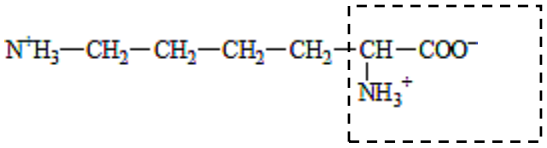
Arginine



Glutamic acid



Histidine



Lysine

Classification of amino acids

Amino acids are classified in several ways depending on the feature on which they are divided into groups. The following classifications of amino acids are accepted:

I. Structural - on the structure of the side radical:

1. Acyclic amino acids

Aliphatic unsubstituted aminoacids (monoamino monocarboxylic) - glycine,alanine,valine,leucine,isoleucine.

Aliphatic substituted amino acids

- Hydroxyamino acids (serine, threonine).
- Sulfur-containing (methionine, cysteine).
- Carboxyamino acids (monoaminodicarboxylic acids) - aspartic acid, glutamic acid.
- Amides of dicarboxylic acids (asparagine, glutamine).
- Diamino acids (diaminomonocarboxylic acids) - lysine, arginine.

2. Cyclic amino acids

- *Aromatic amino acids (phenylalanine, tyrosine).*
- *Heterocyclic amino acids (tryptophan, histidine).*
- *Cyclic imino acid (proline).*

II. By the polarity of radicals

That is, the ability of amino acids to interact with water at physiological values of pH (about pH 7.0):

- **Nonpolar (hydrophobic)** (alanine, valine, leucine, isoleucine, proline, phenylalanine, tryptophan, methionine).
- **Polar uncharged** (glycine, serine, threonine, tyrosine, cysteine, asparagine, glutamine).
- **Polar negatively charged** (glutamic acid, aspartic acid).
- **Polar positively charged** (lysine, arginine, histidine).

III. By acid-base properties:

- **Acidic** - with additional carboxyl groups in the side radical (monoaminodicarboxylic acids: aspartic and glutamic).
- **Alkaline** are diaminomonocarbon (lysine, arginine) and histidine.
- **Neutral** - the rest of the amino acids in which the side radical shows neither acidic nor basic properties.

IV. Biological (physiological) - as amino acids are essential for the body:

Non-essential amino acids. Among the 20 proteinogenic amino acids, 8 amino acids in the human body are synthesized in sufficient quantities de novo from other intermediates (alanine, aspartic acid, aspartic acid, glutamic acid, glutamine, proline, glycine, serine).

Essential amino acids are not synthesized by the enzyme systems of the human body, so they must be supplied with food (valine, leucine, isoleucine, phenylalanine, tryptophan, methionine, threonine, lysine).

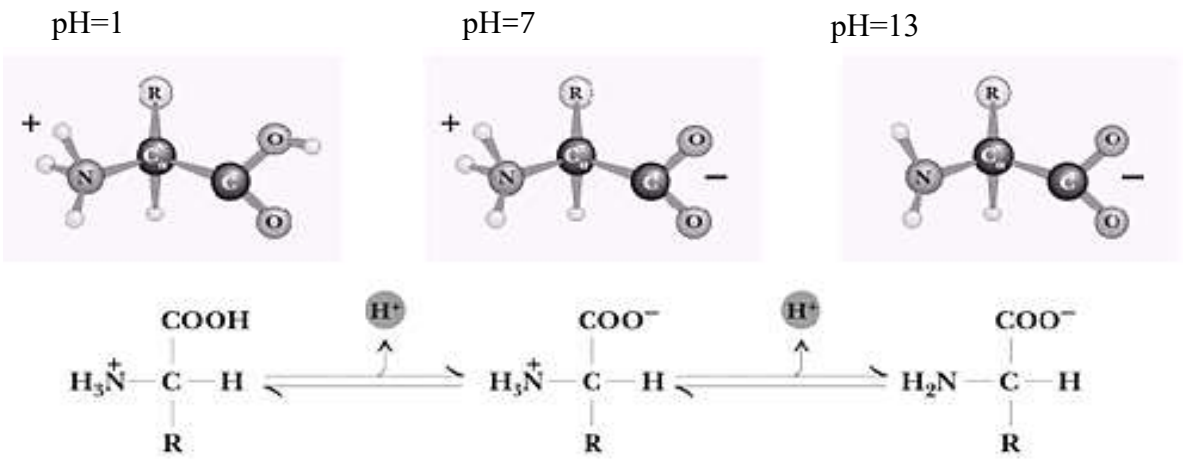
Partially non-essential amino acids - are synthesized in the body in insufficient quantities (arginine, histidine).

Conditionally essential amino acids are synthesized in the body from essential amino acids: cysteine (synthesized from the essential amino acid methionine), tyrosine (synthesized from the essential amino acid phenylalanine).

Physicochemical properties of proteinogenic amino acids

Acid-base properties.

Amino acids have a radical and two functional groups with opposite properties: acid - carboxyl and basic - amino group. Amino acids are amphoteric electrolytes that can dissociate to form ionic forms - anion or cation. In the aqueous medium, amino acids exist in the form of an equilibrium mixture consisting of anionic, cationic forms and bipolar ion (zwitter-ion).



cationic form
total charge “+1”

bipolar form (electroneutral)
total charge “0”

anionic form
total charge “-1”

Methods for detection and study of amino acids in biologicalliquids

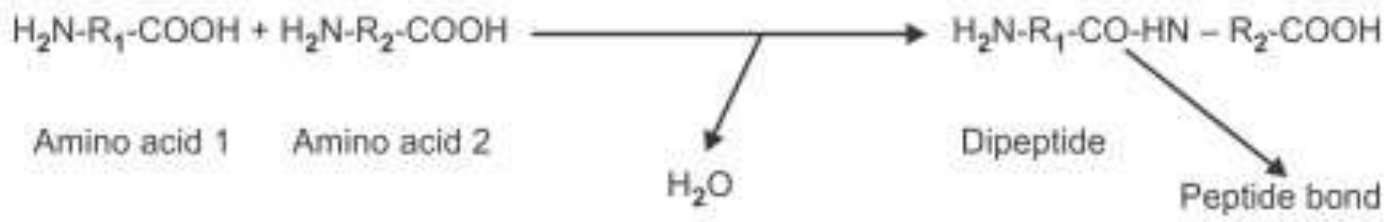
Amino acids color reactions:

- ***Ninhydrin reaction.***
- ***Fluorescamine reaction.***
- ***Xanthoprotein reaction.***
- ***Milo's reaction.***
- ***Sakaguchi's reaction.***
- ***Ehrlich's reaction.***
- ***Fol's reaction.***

Natural peptides: classification, biochemical characteristics

The human body produces many peptides that are involved in the regulation of various biological processes and have high physiological activity. The number of amino acid residues in the structure of biologically active peptides can vary from 3 to 50. The smallest size of peptides can include thyrotropin-releasing hormone and glutathione (tripeptides), as well as enkephalins, which contain 5 amino acids. However, most biologically active peptides contain more than 10 amino acids, for example, neuropeptide Y (appetite regulator) contains 36 amino acids, and corticoliberin - 41 amino acids.

The amino acids are joined by peptide bonds in proteins. The dipeptide formation from two amino acids occurs with a loss of a water molecule. If amino acid 1 is glycine and 2 is alanine, then the dipeptide formed is glycylalanine. If the amino acids are interchanged, then the resulting peptide is alanylglycine. Always the amino group of the first amino acid in the peptide is free and the carboxyl group of the last amino acid is also free. The amino terminal of the peptide is always written on the left side and carboxyl terminal will be on the right side.



Currently discovered and studied peptides can be classified into groups according to their main physiological action:

- **peptides with hormonal activity** (oxytocin, vasopressin, hypothalamic releasing hormones, melanocyte-stimulating hormone, glucagon, etc.);
- **peptides that regulate digestive processes** (gastrin, cholecystokinin, vasointestinal peptide, etc.);
- **peptides that regulate vascular tone and blood pressure** (bradykinin, kalidin, angiotensin II);
- **appetite-regulating peptides** (leptin, neuropeptide Y, melanocyte-stimulating hormone, β-endorphins);
- **peptides that have an analgesic effect** (enkephalins and endorphins and other opioid peptides). The analgesic effect of these peptides is hundreds of times greater than the analgesic effect of morphine;
- **peptides involved in the regulation of higher nervous activity**, in biochemical processes associated with the mechanisms of sleep, learning, memory, fear, etc .;
- **peptides-antioxidants** (glutathione).

Proteins

Proteins are high molecular weight natural polymers consisting of amino acid residues linked by a peptide bond; is the main component of living organisms and the molecular basis of life processes.

Classification of proteins.

I. Functional

Depending on the function that proteins perform in the body. There are catalytic, contractile, structural, transport, protective, regulatory, receptor proteins.

II. By shape

1. **Globular proteins** are compact spherical molecules, water-soluble (the ratio of molecule length to diameter does not exceed 4); globular proteins perform dynamic functions (enzymes, immunoglobulins and transport proteins hemoglobin and albumin).

2. **Fibrillar proteins** have a rod-shaped elongated shape, insoluble in water (the ratio of the length of the molecule to a diameter greater than 10); they perform mainly structural and protective functions (eg, collagen).

III. According to the degree of complexity of the molecule

1. Simple proteins (consisting only of amino acids).

The classification of **simple proteins** is based on solubility. Simple proteins include:

- *Albumins*
- *Globulins*
- *Protamines*
- *Histones*
- *Prolamines*
- *Glutelins*
- *Scleroproteins*

2. Complex proteins (consisting of a simple protein bound to a non-protein component).

The classification of complex proteins is based on the structure of the non-protein component. Complex proteins include:

- *Chromoproteins*
  - *1) Hemoproteins*
  - *2) Flavoproteins*
- *Glycoproteins*
- *Lipoproteins*
- *Metalloproteins*
- *Nucleoproteins*
- *Phosphoproteins*

Physico-chemical properties of proteins

Individual proteins differ in their physicochemical properties: the shape of molecules, molecular weight, total charge of the molecule, the ratio of polar and nonpolar groups on the surface of the native protein molecule, solubility, and the degree of resistance to denaturing agents.

1. Differences in the shape of proteins.

According to the shape of molecules, proteins are classified into globular and fibrillar. Globular proteins have a more compact structure, their hydrophobic radicals are hidden in the hydrophobic nucleus, and they are much more soluble in body fluids than fibrillar proteins (with the exception of membrane proteins).

2. Differences in protein molecular weight.

Proteins are macromolecular compounds, but can vary greatly in molecular weight, ranging from 6,000 to 1,000,000 Da and above. The molecular weight of a protein depends on the number of amino acid residues in the polypeptide chain, and for oligomeric proteins - on the number of protomers (or subunits) that are part of it.

3. Total charge of proteins.

Due to the electrolytic dissociation of NH_2 and COOH groups, proteins exhibit the properties of amphoteric compounds. Proteins contain radicals of lysine, arginine, histidine, glutamic and aspartic acids, which contain functional groups capable of ionization (ionic groups). In addition, at the N- and C-terminus of the polypeptide chains there are α-amino and α-carboxyl groups, also capable of ionization. The total charge of the protein molecule depends on the ratio of ionized anionic radicals Glu and Asp and cationic radicals Lyz, Arg and His.

4. The ratio of polar and non-polar groups on the surface of native protein molecules.

Polar radicals predominate on the surface of most intracellular proteins, but the ratio of polar and nonpolar groups differs for individual proteins. Thus, the protomers of oligomeric proteins in contact with each other often contain hydrophobic radicals. The surfaces of proteins that function in membranes or attach to them during operation are also enriched with hydrophobic radicals. Such proteins are better soluble in lipids than in water.

5. Solubility of proteins.

The solubility of proteins in water depends on all the above properties of proteins: shape, molecular weight, magnitude of charge, the ratio of polar and non-polar functional groups on the surface of the protein. In addition, the solubility of the protein is determined by the composition of the solvent, ie the presence in the solution of other solutes. For example, some proteins are more readily soluble in weak saline than in distilled water. On the other hand, increasing the concentration of neutral salts can contribute to the precipitation of certain proteins in the sediment (salting).

6. Differences in proteins by the degree of resistance to denaturing agents.

Denaturing agents present in the solution also reduce the solubility of proteins. The influence of denaturing factors is used for sterilization of equipment and instruments, as well as as antiseptics.

Denaturation - a violation of the spatial structure of the protein (loss of secondary, tertiary and Quaternary (if any) structures while maintaining the primary structure) and violation of the characteristic physical and chemical properties of this protein. Denaturation is accompanied by a decrease or loss of specific protein-specific biological activity (enzymatic, hormonal, etc.). The mechanism of action of denaturing agents is the destruction of weak bonds (hydrogen, ionic, dipole, hydrophobic), which stabilize the secondary, tertiary and Quaternary structures of protein molecules. The primary structure is preserved because it is formed by strong covalent bonds. Denaturation can be reversible (the structure of the protein is restored after the removal of the denaturing agent - renaturation) or irreversible (the spatial structure of the molecule is not restored).

Factors that cause protein denaturation can be divided into physical and chemical.

Physical factors include:

•high temperature;
•ultraviolet radiation;
•X-ray and radioactive irradiation;
•ultrasound;
•mechanical impact.

Chemical factors include:

•concentrated acids and alkalis. For example, trichloroacetic acid (organic), nitric acid (inorganic);
•heavy metal salts (eg $CuSO_4$);
•organic solvents (ethyl alcohol, acetone);
•plant alkaloids;
•urea in high concentrations;
•other substances that can break weak types of bonds in protein molecules.

Levels of structural organization of protein molecules

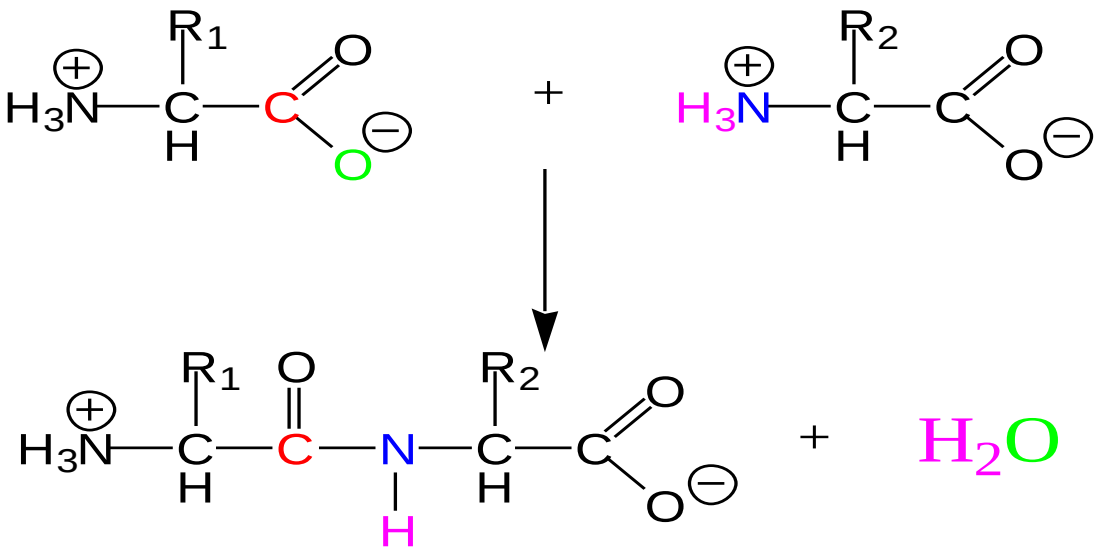
In addition to the amino acid sequence of the polypeptide, the three-dimensional structure formed during folding is extremely important for the functioning of proteins. This structure is maintained as a result of the interaction of lower-level structures. The three-dimensional structure of proteins under normal natural conditions is called the native state of the protein. Loss of the native state is denaturation.

There are *four levels* of structural organization of proteins:

- **Primary structure**
- **Secondary structure**
- **Tertiary structure**
- **Quaternary structure**

Primary Structure

This is a polypeptide chain configuration that is formed by the formation of an acid amide (**peptide**) bond between amino acid residues. In the synthesis of the peptide, the **α-carboxyl group** of one amino acid interacts with the **α-amino group** of the second amino acid, forming a peptide bond with the release of a water molecule:



When two amino acids interact, a dipeptide is formed, three - a *tripeptide*, and so on until the formation of a huge polypeptide. It is conventionally accepted that peptides containing from 2 to 20 amino acid residues belong to *oligopeptides*; those having from 20 to 50 amino acid residues - to *polypeptides*. Peptide chains, which combine more than 50 amino acids and have a molecular weight greater than 6,000, belong to the actual proteins.

The primary structure of each individual protein is encoded in the DNA molecule and is realized during transcription (transmission of information encoded in DNA to mRNA molecules) and translation (protein synthesis).

Peptide bonds are very strong, and harsh conditions are used for their chemical non-enzymatic hydrolysis. In a living cell, peptide bonds can be broken by proteolytic enzymes called proteases or peptide hydrolases.

The primary structure of the protein is stabilized:

- **peptide bonds** (between amino acid residues);
- **disulfide bonds** (between free -SH-groups of cysteine).

The primary structure of a protein carries information about its spatial structure.

Secondary Structure

The secondary structure of proteins is a local conformation caused by the rotation of individual sections of the polypeptide chain around single covalent bonds.
The main bonds that stabilize the secondary structure are *hydrogen* bonds between atoms of peptide groups of amino acids in the same peptide chain or between different peptide chains.

Types of secondary structure:

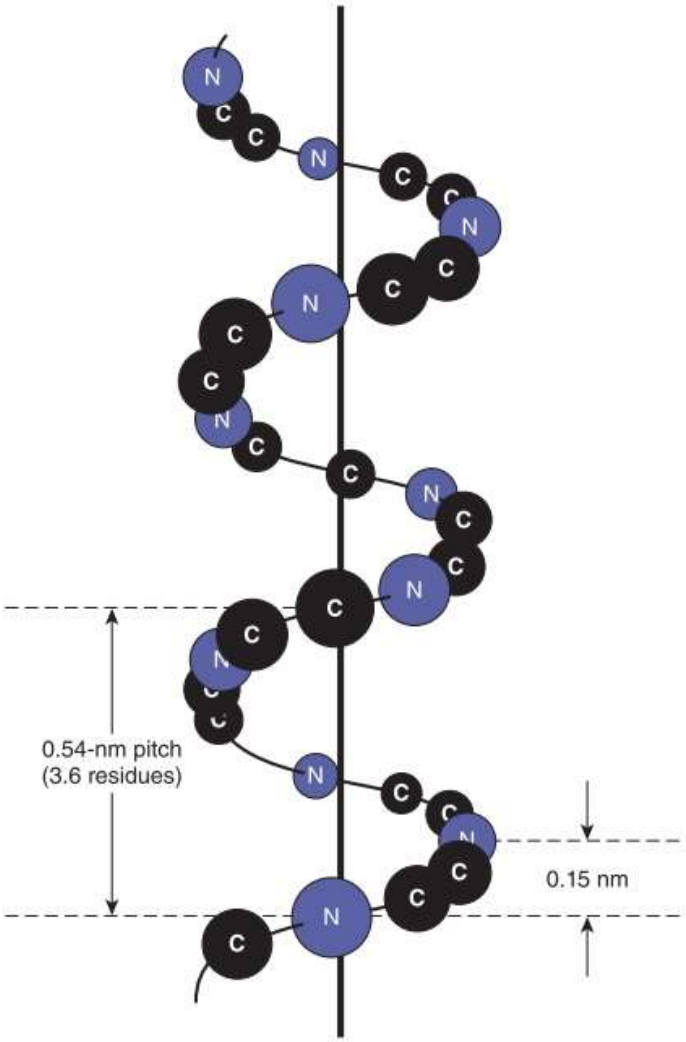
- **α-helix (right-handed)**
- **β-pleated sheet**

Alpha helix

In the α-helix, hydrogen bonds are formed between the oxygen atom of the carbonyl group and the hydrogen amide nitrogen of the 4th amino acid.

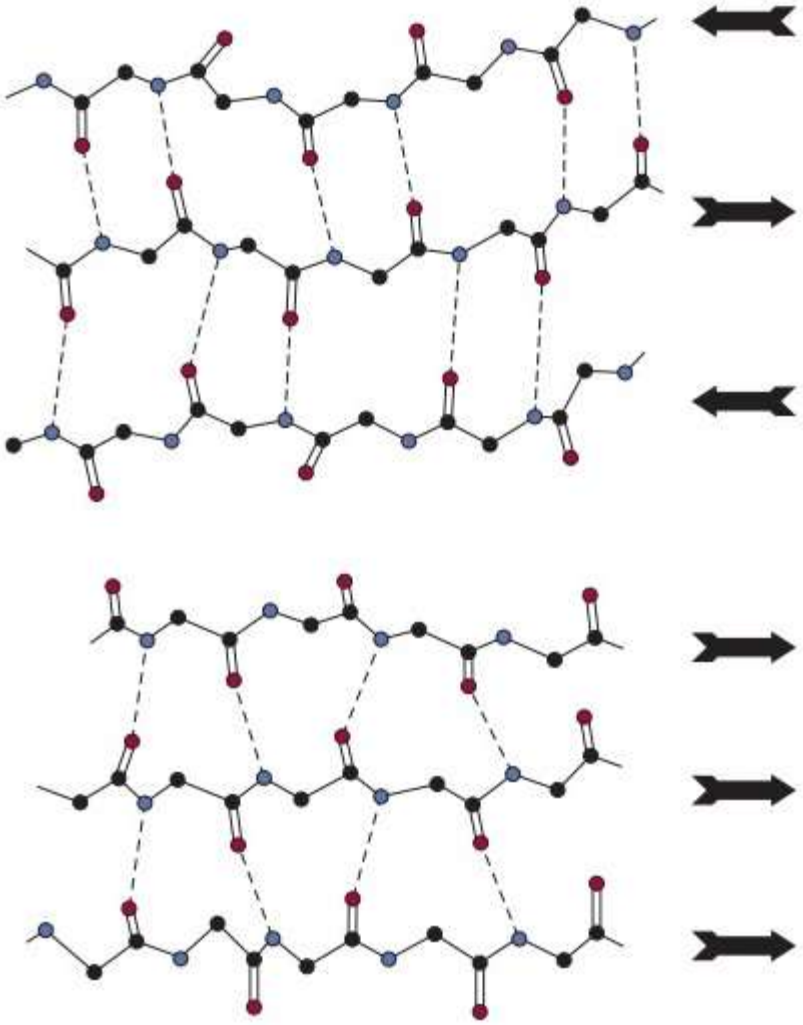
Hydrogen bonds are located parallel to the axis of the spiral and are repeated many times, so they hold the spiral structure firmly in a slightly tense state (like a compressed spring).

Lateral radicals of amino acid residues are located on the periphery of the helix and do not participate in the formation of the secondary structure. The helix pitch (one complete turn) of 0.54 nm is 3.6 amino acid residues per turn, and one amino acid residue is 0.15 nm.



Beta sheet

The β-structure is formed between the linear regions of the peptide framework of one polypeptide chain, forming folded structures. The β-structure is stabilized by hydrogen bonds between C = O and NH-groups. Polypeptide chains or parts thereof can form **antiparallel (← → ←)** or parallel (→→→) β-structures. In the antiparallel folded sheet, the chains are directed in opposite directions. In a parallel folded sheet, both peptide chains have the same direction in space. Both structures occur in natural proteins and peptides, but antiparallel is more stable and therefore more common.



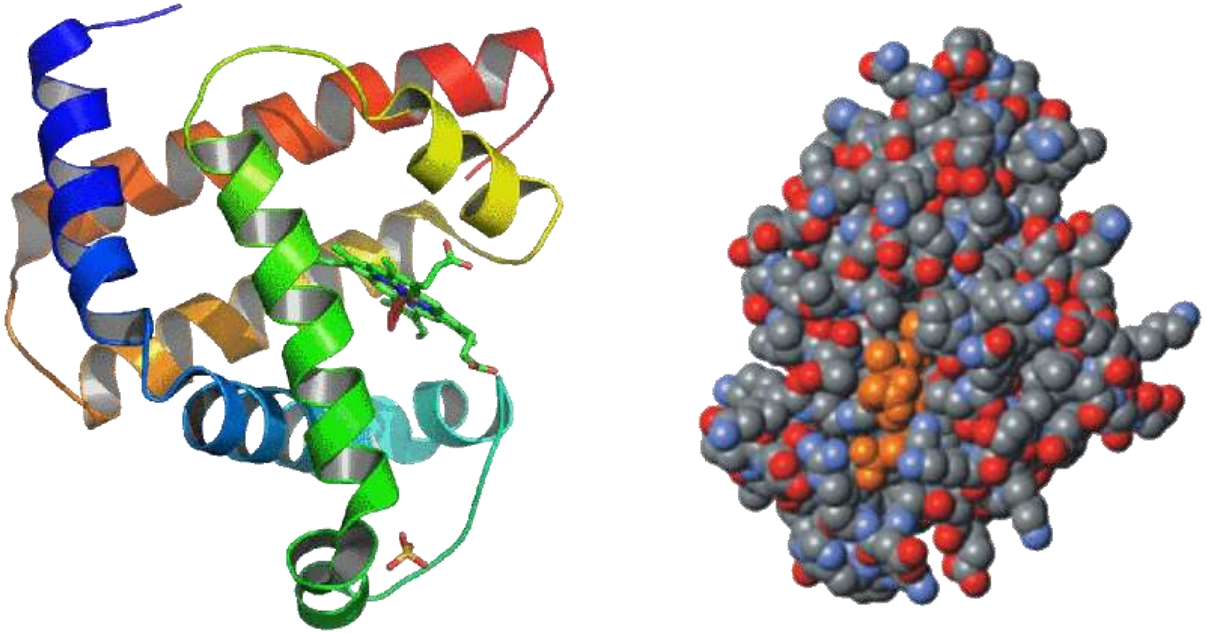
Tertiary structure

The tertiary structure of proteins is a three-dimensional spatial structure formed by interactions between amino acid radicals, which can be located at a considerable distance from each other in the polypeptide chain. Connections involved in the formation of the tertiary structure of proteins:

1) hydrophobic interactions. When laying the polypeptide chain of the protein tends to take an energetically beneficial form, characterized by a minimum of free energy. Therefore, hydrophobic amino acid radicals tend to combine within the globular structure of water-soluble proteins. Between them there are so-called hydrophobic interactions, as well as van der Waals forces between closely adjacent atoms. As a result, a hydrophobic nucleus is formed inside the protein globule;ядро;

2) ionic bonds. Ionic bonds can occur between negatively charged (anionic) carboxyl groups of aspartic and glutamic acid radicals and positively charged (cationic) groups of lysine, arginine or histidine radicals;

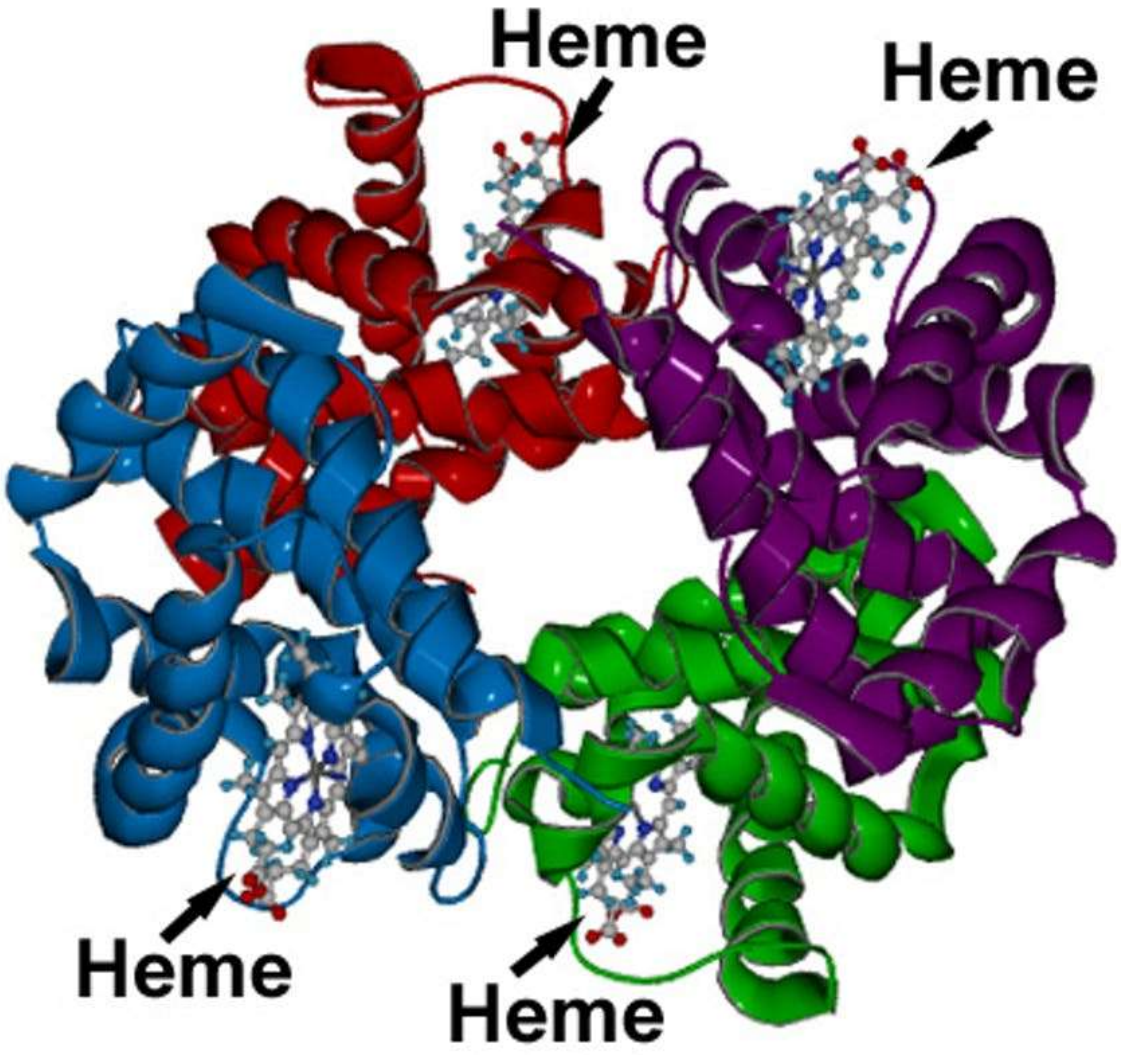
3) hydrogen bonds. Hydrophilic amino acid radicals tend to form hydrogen bonds with water and are therefore mainly located on the surface of the protein molecule. All hydrophilic groups of amino acid radicals inside the hydrophobic nucleus interact with each other through ionic and hydrogen bonds.



Quaternary structure

A quaternary structure is formed by combining several polypeptide chains having a tertiary structure.
The protein thus formed has a new function. Proteins with a Quaternary structure are called oligomeric, and the individual polypeptide chains that form them are called protomers or monomers. Such protomer combinations are stabilized by weak non-covalent bonds (hydrogen, hydrophobic, electrostatic interactions) between amino acid residues located on the surface of the protomers.
An example of a protein with a Quaternary structure is hemoglobin. Its molecule is made up of four identical subunits - two α- and two β-polypeptide chains, each of which is connected to a non-protein compound heme - a porphyrin derivative that binds the oxygen molecule.

The structure of hemoglobin



Biological functions of proteins and peptides

1. Catalytic function. More than 3400 proteins are enzymes. Enzymes - a type of protein characterized by specific catalytic properties, ie each enzyme catalyzes one or more reactions. Enzymes catalyze the reactions of cleavage (catabolism) and synthesis (anabolism) of complex molecules, in particular, the synthesis and degradation of DNA, RNA, proteins, lipids and sugars. In addition, they catalyze the synthesis and degradation of small molecules, chemical modifications and a number of other reactions necessary for life.

2. Contractile function. Proteins are involved in mechanical contraction for movement (usually have adenosine triphosphatase activity): actin and myosin of muscles, proteins of flagella and cilia of protozoa, flagella of sperm, tubules of the chromosome movement during mitosis, etc.

3. Structural function. Structural proteins often play the role of reinforcement, giving shape and rigidity to cells and tissues. Usually these proteins are able to form long filaments or bind filaments formed by other proteins - some structural proteins are fibrillar, others form filaments by polymerization of protein globules under certain conditions. The main structural proteins are collagen, elastin (forming the bone matrix, vascular system, and other organs), α-keratin (present in epidermal tissue), plasma membrane proteins, and others.

4. Protective function. Many proteins in the blood are involved in the body's protective response to both damage and attack by pathogens. Proteins that perform this function include antibodies (immunoglobulins), which are produced in response to the introduction of antigens; proteins of the blood coagulation system; complement system proteins; xenobiotic neutralizing enzymes; interferons, interleukins, lysozyme, etc.

5. Transport function. Proteins bind and transport in the blood and cell of various ligands - biomolecules, metal ions, foreign chemical compounds (xenobiotics). Examples of transport proteins are serum albumin (carries fatty acids, bilirubin, drugs and toxic compounds), erythrocyte hemoglobin (transports oxygen), lipoproteins (transports lipids), transferrin (transports iron), and others.

6. Regulatory function. Numerous bioregulators have a protein and peptide nature - hormones, mediators and modulators produced in the endocrine system, brain neurons, immune system: simple proteins (insulin, somatotropin, prolactin, etc.), glycoproteins (thyrotropin, lyonimonut, folliculost). low molecular weight peptides (vasopressin, oxytocin, opioid brain peptides, etc.). Examples of regulatory proteins are also histones, which stabilize the structure of DNA and regulate the functioning of the genome; heat shock proteins (stress proteins); G-proteins that regulate the synthesis of cyclic nucleotides; oncoproteins and antioncoproteins that determine cell malignancy.

7. Participation in maintaining blood pH. Buffer properties, which are due to the presence of free amino and carboxyl groups in the protein structure. (hemoglobin and protein buffer systems).

8. Receptor function. Receptor proteins in the internal environment of the body are used to interact with molecules-bioregulators (signaling molecules). Localized in the membrane structures of cells, and can be in a dissolved state. Thus, membrane receptors for physiologically active compounds that receive a chemical signal from hormones, neurotransmitters (adrenoceptors, cholinoreceptors, histamine receptors, etc.) have a protein nature. Photoreceptor proteins (opsin) are known for the perception of signals from the environment, cholinoreceptor proteins and the like for sound perception.

9. Maintaining oncotic pressure in cells and blood (albumin) thus involved in the regulation of water metabolism between blood and extracellular space.

10. Energy function. (very little, because the products of protein hydrolysis are a source of energy only in special conditions, such as starvation).

11. Plastic function. In case of protein deficiency, albumin can be used by tissues as a plastic material to build their own proteins.

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