Proteins

Definition

Macromolecular compounds built exclusively or in major range from aminoacids, which are the largest part of organic compounds found in living cells.

- Do not cross dialysis membranes
- Undergo denaturation (damage to secondary structures)
- Molecular weight above 10 000 D

Proteins

Divisions

- in accordance to origin – plant and animal
- in accodance to the presence in organs – plasma, muscles, milk, ribosomal, cellular, microsomal
- in accordance to biological function – enzymatic, transport, receptor
- in accordance to structure – single – contain only aminoacids
  - conjugated – contain additional compounds
- in accordance to the differences in solubility and the shape of molecule
  - globular
  - fibrous

Physicochemical properties of proteins

- colloidal solutions (sols ↔ gels)
- diffusion
- the lack of ability to dialysis
- well solubilised in water
- electric charge (pI)
- denaturation (temp, acids, bases, alkohol, aceton, cations of heavy metals, urea, guanidin hydrochloride)

Proteins

The meaning of proteins

- active – enzymes, hormons
- **structural** – connective tissue (stiffening, motility), bones (elasticity) – collagen, keratin, elastin
- immune – immunoglobulins
- clotting factors
- **transport** – transport electrons, metabolic products and gases – hemoglobin, ceruloplasmin, transferrin
- storage – ferritin, mioglobin
- **contractile** – actin, myosin, troponins
- **receptor** – mediate in specific binding of biologically active substances in place of action

Fibrous proteins

- they have structural functions
- they are sparingly soluble in water and diluted salt solutions (exception is fibrynogen in blood plasma)
- they have high mechanical strength
- they are characterised by the parallel arrangement of polypeptides in forms of long fibrils forming structural elements
- we include here: keratins (hair, wool)
  collagens (cartilage, skin)
  elastins (tendons, ligaments)

Globular proteins

- they have the shape of a sphere
- they are well soluble in water
- the compact structure results from the specific rippling of polypeptide chain, caused by hydrophobic interactions between non-polar aminoacid residues
- they are stabilized by hydrophobic and hydrophilic interactions
- we include here: mioglobin (α-helix, oxygen storage in muscles)
  lysozyme (α and β structure)
  immunoglobulins (β structure)
  all enzymes, blood proteins (excluding fibrynogen)

Protein structures

PRIMARY – determines the number and order (sequence) bound by peptide bonds aminoacids in chain, genetically conditioned

SECONDARY – describes the conformation of chains resulting from the formation of hydrogen bonds between carbonyl and amide group of the main polypeptide chain (helical and pleated structure), conditioned by primary structure

The properties of protein α-helix

- structure of α-helix is stabilized by **hydrogen** bonds which are formatted between H atom of –NH- and O atom of carbonyl group of 1st and 4th peptide bond in the same chain
- each peptide bond participates in formatting of hydrogen bonds what ensures high durability (1→4; 3.6 aminoacid residues)
- the right-handed helix is more stable that left-handed
- α-helix arises spontaneously because this conformation is more stable for polypeptide chain

β pleated sheet

Hydrogen bonds connect adjacent parallel and antiparallel polypeptide chains

β turns

When β turn takes place the chain bends 180^0.

Most often it is caused by the presence of prolin.

Protein structures

TERTIARY – defines the three dimensional corrugation of a given polypeptide chain caused by intramolecular interaction of aminoacid side chains

QUATERNARY – covers the association between two or more polipeptide chains in certain molecular complexes

Bonds in protein structures

- hydrogen bonds – are created between peptide bonds of the same or adjacent chain. They are not strong but due to their number play a crucial role in the stablization of protein structure.
- disulfide bonds (covalent) – are created between two residues of cystein of the same or adjacent chains
- ionic bonds – are created between additional amino and carboxyl groups of the same or adjacent chains
- hydrophobic bonds – the weakest – mutual interactions between apolar side chains of aminoacids

Structure – function dependence of selected proteins

Collagen

Collagen is the main extracellular structural protein, the component of connective tissue. Polysaccharide units bound via O-glycosidic bond with hydroxylysine may appear. Collagen is mechanically strong but flexible.

It is not soluble in water or electrolyte solutions. This results from cross-linking by covalent lateral bonds.

Main aminoacids: glycine 33%, proline 10%, hydroxyproline 10%, hydroxylysine 1% – α-helix cannot be created. The lack of cysteine and tryptophan.

Structure of young collagen is maintained by non-covalent bonds (hydrophobic and electrostatic interactions, hydrogen bonds). Together with aging of protein more and more covalent cross-linking bonds (lateral) are created - mainly intra and inter molecular bonds between side chains of lysin and hydroxylysin.

Collagen structure

Tropocollagen is right-handed superhelix consisting of 3 polypeptide left-handed helices (285 kDa) – two identical α1 and one α2, each contains around 1000 of aminoacid residues. Every third aminoacid is glycine. The structure is much more extended as α helix. The structure is difficult to unravel.

The alterations in collagen metabolism

Scurvy – insufficiency of vitamin C (cofactor of enzymatic reactions that catalyse the hydroxylation of proline and lysine) may lead to disturbances in the formation of collagen

Symptoms: abnormal bone development, easy bruising and bleeding due to fragile blood vessels, tooth loss, slow wound recovering, osteoporosis

Prophylaxis and treatment – daily intake of vitamin C in the diet

Osteogenesis imperfecta (excesive bone fragility – genetic defect (mutation of gene leading to the disturbances in the synthesis of collagen chains and not stable collagen superhelix)

Symptoms: weakness of bones and their deformation

Treatment: orthopedic corrections

Ehlers-Danlos syndrome – genetic defect (mutation of gene leading to abnormalities in collagen synthesis)

Symptoms: weakness of connective tissue, excessive stretch of skin and joints, loosening of musculo-skeletal connections

Treatment: none, sometimes vitamin C can be prescribed

Keratins

Two α helix chains are subjected to dextrorotary folding and then with the same unit they also curl left-handedly (protofibrils – disulfide bonds). 8 protofibrils create mikrofibril, while several hundred - macrofibril.

Keratins

During stretching the structural form of α keratin may form stretched β keratin.

The ordered structure of keratin molecules is maintained by numerous hydrogen, disulfide and hydrophobic bonds.

These proteins are characterised by high content of cystein and aminoacids with non-polar side chains.

Elastin

Elastin is the main component of tendons, muscular ligaments and blood vessel walls, usually accompany collagen.

Tropoelastin molecule consists of around 860 aminoacid residues. Over 80% of them are non-polar aminoacids, 30% of which is glycine and about 60% proline and alanine.

Mature elastin contains many covalent cross-links formed by use of desmosin (derivative of lysine).

Elastin is water insoluble, does not convert into gelatin during boiling, does not undergo hydrolysis under the influence of digestive enzymes, hydrolyze under the action of elastase.

Albumins

Albumins are synthesised in liver and constitute 55-60% of total proteins in plasma.

The ellipsoid shape of the molecule does not increase the viscosity of blood and is easily transported in the bloodstream.

Albumin is very acidic protein with large negative charge in pH conditions of blood. It helps to bind molecules of cations and increases their possibility to transport water from intercellular space into bloodstream.

Albumin contains at least 3 domains binding different molecules

Albumins

1. Transport

- metal ions – calcium and cooper
- free fatty acids
- bilirubin
- salts of bile acids
- hormones (thyroid glands and steroid)

2. Maintenance of osmotic pressure (oncotic)

Albumins

Albumins bind many drugs and influence their effectiveness

Sulfonamids via competitive binding cause the liberation of bilirubin from albumins

The decrease in albumin synthesis appears during starvation and liver diseases. It may lead to oedema and retention of water in tissues.

Gamma globulins

Specific structure of antibodies allows for the recognition and binding antigens what causes immunological answer.

Functional domains:

- Binding domain
- Effector domain

Muscle fibers

- **Thick filaments - myosin**
- **Thin filaments – actin, tropomyosin, troponin**

The structure of filaments is specific –thick are bipolar, antiparallel thin which surrounds thick slip along thick filaments, shortening the sarcomeres.

Haemoglobin

- it is a tetramere of non-covalently bound subunits, each contains heme which is the center for binding oxygen

- it is able to bind oxygen at high P_{O2} in lungs and to liberate it in tissues at low P_{O2} . Similarly it has to bind CO2 from tissues and liberate in lungs.

Haemoglobin

- Mechanism of cooperative binding of oxygen

In deoxyhaemoglobin iron lies outside the heme plane. After oxygenation atom shifts and allows for further binding of oxygen – the change in conformation from T into R (oxy).

Haemoglobin

- allosteric effectors: protons (H+), CO2, 2,3 bisphosphoglycerate
- at the increase of pH and P_{CO2} oxygen binding affinity decreases – in tissues
- binding of protons causes the shift from conformation R into T, CO2 binding increases
- in lungs at pH 7.4 and high P_{O2} – shift from conformation T into R
- the increase in 2,3 DPG supports the shift to T conformation and the liberation of oxygen in tissues

Haemoglobin – clinical aspects

- Glycosylation of haemoglobin – monitoring of glucose concentration

- Haemoglobinopathies – genetic diseases resulted from mutations in haemoglobin

- Talasemie – haemolytic anaemia resulted from insufficient production of hemoglobin units

Glycoproteins or proteoglycans?

Glycoproteins

Glycoproteins are proteins containing covalently bound oligosaccharides (2-10 residues of N-acetylhexosamin) which are added during posttranslational modifications of proteins. Often act as biologically active molecules such as hormones or enzymes. They protect from proteolysis and are responsible for the recognition of antigenes. They are the main component of blood groups and many receptors located on the surface of cells.

Proteoglycans

Proteoglycans are macromolecular components of extracellular matrix consisting of protein medulla – around 6-20% of molecule which is covalently bound to glycosaminoglycan chains (80-94%). As the members of extracellular matrix they bind polycations (they are polyanions due to the presence of sulfur groups carrying negative charge) to assure appropriate hydratation of connective tissue and its turgor.

Hyaluronic acid due to the lack of ability to create covalent bonds with proteins and the lack of sulphur groups is not included into proteoglycans.

Lipoproteins or proteolipids?

Lipoproteins

Protein-lipid complexes connected by non-covalent bonds which are present in blood plasma.

- Chylomicrons
- VLDL
- LDL
- HDL

Proteolipids

Molecules built from proteins and covalently bound cholesterol, glycophosphatydyloinositol and fatty acids.

They participate in the process of cell signalling, the stabilisation of protein conformation and apoptosis.