Enzyme preparations.

General characteristics: definition, specificity of enzymatic reactions. Classification and nomenclature of enzymes. Enzyme preparations of plant and animal origin. Immobilized enzymes. Immobilization methods. Water-soluble preparations of immobilized enzymes. Incorporation of enzymes into microcapsules. Incorporation of enzymes into liposomes. Preparations of immobilized enzymes used in local diseases. Standardization of enzyme preparations. Methods for assessing enzymatic activity.

Private technology.

LECTURE PLAN

1. Enzyme drugs. General characteristics: definition, specificity of enzymatic reactions.Enzyme stability.

2. Classification and the nomenclature of enzymes.

3. Standardization of enzyme preparations. Methods for assessing enzymatic activity.

4. Enzyme preparations of plant and animal origin.

5. Immobilized enzymes. Immobilization methods. Water-soluble preparations of immobilized enzymes. Incorporation of enzymes into microcapsules. Incorporation of enzymes into liposomes.

6.Preparations immobilized enzymes used for local diseases.

7. Private technology.

Basic concepts

- **Enzymes** (lat. *fermentum* - "leaven") - biologically active substances of a protein nature, accelerating various chemical transformations of substances – **substrates**.

Term fermentum was proposed in XVIII by Van-Helmont for substances affecting on alcoholic fermentation

- **Enzymes** (Greek. *en zyme* - "in yeast") Is a synonym for the word “enzymes”.
- **Enzymology** - a section of biochemistry that studies enzymes.

Enzymes as proteins

- consist of many hundreds of amino acid residues connected in a specific sequence using peptide bonds;
- have a globular spatial structure that determines their functionality;
- differ in molecular weight, electrophoretic mobility;
- water-soluble.
- All the features of the structural organization of proteins are inherent in enzymes. When the levels of organization of the protein molecule are violated, the enzyme loses its physicochemical and biological properties
- Violation of the structure of a protein molecule can occur under the influence of:

- physical factors (temperature, pressure, ionizing radiation, mechanical stress, etc.);
- chemical factors (acids, alkalis, organic solvents, heavy metals, etc.);

Enzymes as catalysts

- **Similar to inorganic catalysts:**
  - catalyze energetically possible reactions;
  - increase the speed of both forward and backward reactions;
  - do not change the position of equilibrium;
  - are not consumed during the reaction.
- **Activation energy** Is the energy required to force substances to react.
- Catalysts accelerate chemical reactions by finding workarounds that allow molecules to cross the activation barrier at a lower energy level.

Enzymes help speed up chemical reactions in the human body. They bind to molecules and alter them in specific ways. They are essential for respiration, digesting food, muscle and nerve function, among thousands of other roles.

What do enzymes do?

- **The digestive system** – enzymes help the body break down larger complex molecules into smaller molecules, such as glucose, so that the body can use them as fuel.
- **DNA (**Deoxyribonucleic acid) **replication** – each cell in your body contains DNA. Each time a cell divides, that DNA needs to be copied. Enzymes help in this process by unwinding the DNA coils and copying the information.
- **Liver enzymes** – the liver breaks down toxins in the body. To do this, it uses a range of enzymes.

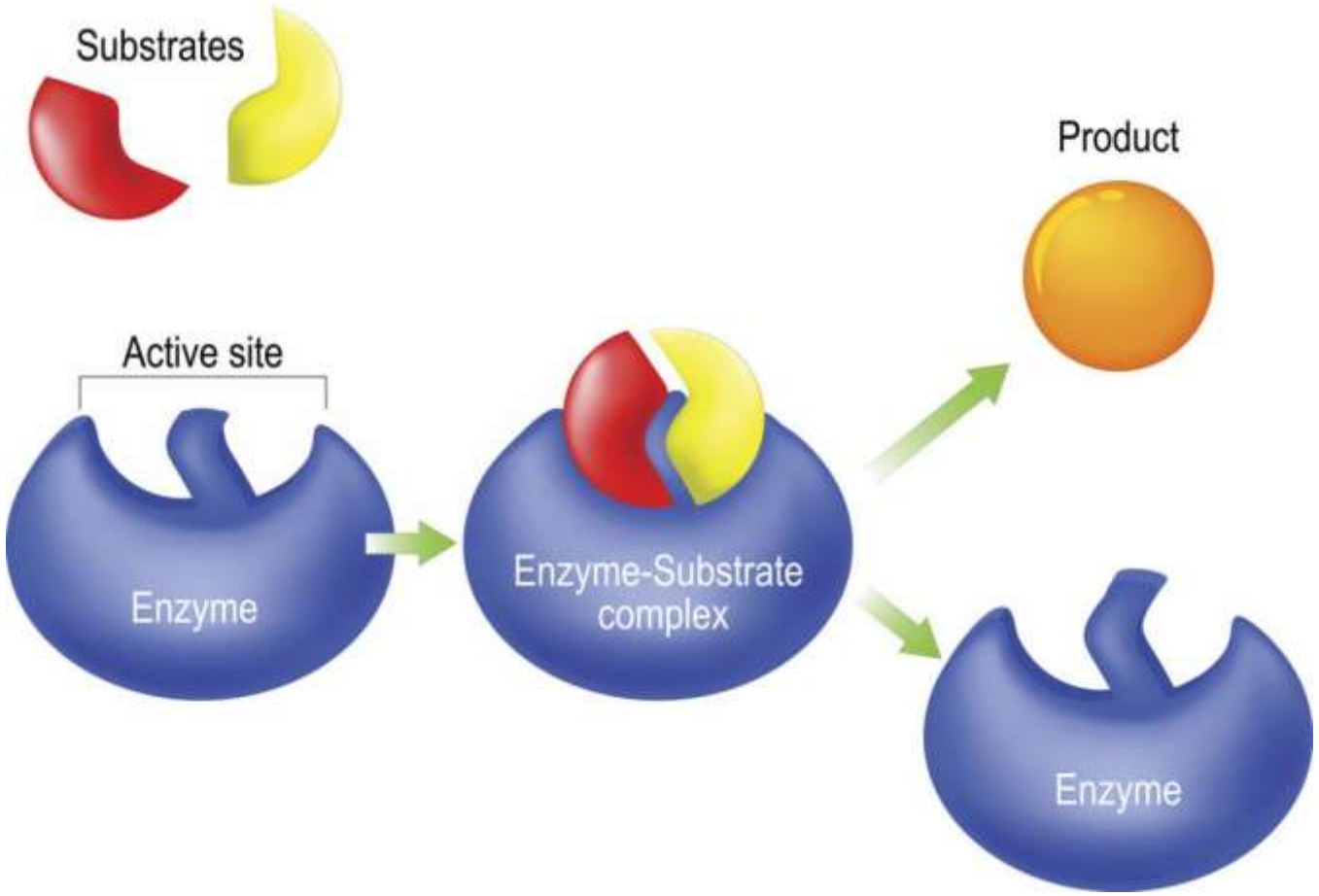
How enzymes work

Enzyme lock and key model

The “lock and key” model was first proposed in 1894. In this model, an enzyme’s active site is a specific shape, and only the substrate will fit into it, like a lock and key.

This model has now been updated and is called the **induced-fit model**.

In this model, the active site changes shape as it interacts with the substrate. Once the substrate is fully locked in and in the exact position, the catalysis can begin.



Structural organization of enzymes

- **Simple enzymes** - simple proteins, they contain only amino acid residues (class 3 enzymes).
- **Complex enzymes** (**holoenzymes)** - complex proteins, they consist of a protein part (**apoenzyme**) and non-protein part (**coenzyme, prosthetic Group**).
- The structure of the apoenzyme determines the specificity of the catalyzed reaction, and the structure of the coenzyme determines its type.

Coenzyme function

- **Coenzymes are carriers of certain atoms, electrons or chemical groups to the corresponding acceptor.**
- In the reaction, the coenzyme undergoes chemical transformations opposite to those that occur in the substrate.
- In subsequent coupled reactions, changes in the coenzyme proceed in the opposite direction.

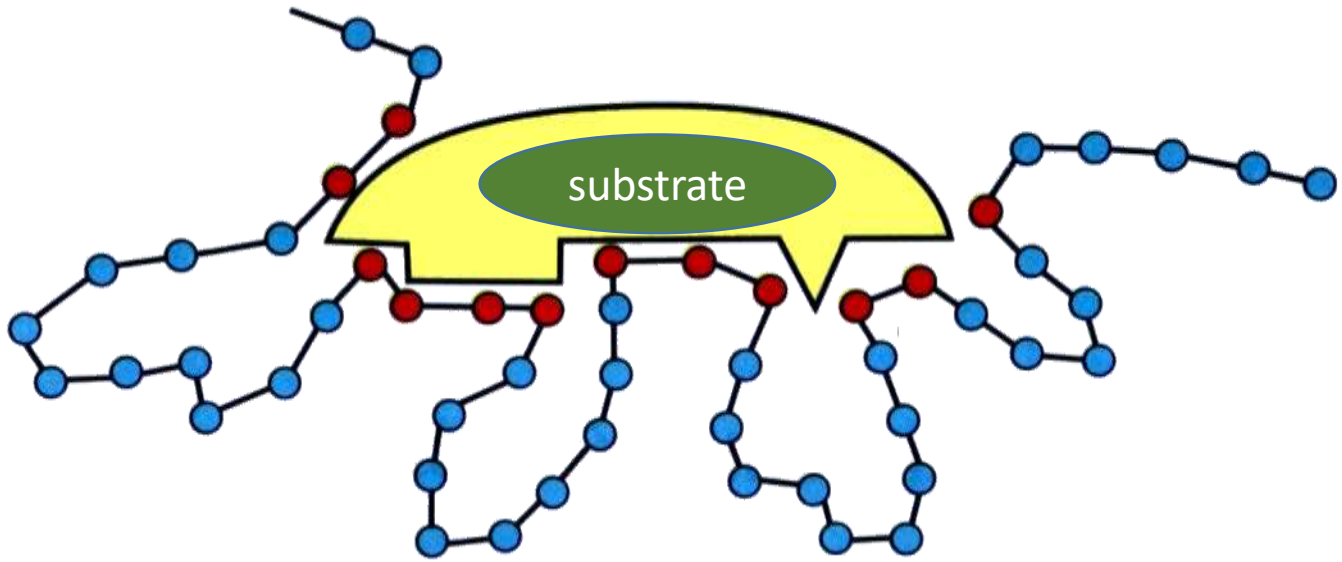
Most often, coenzymes are **vitamins** (derivatives vitamin B_{one} (thiamine), IN_3 (pantothenic acid), vitamin $B_{2,}$ $IN_{6.}$ Vitamin B_{12}, PP, biotin.

Non-vitamin coenzymes: ATP is a coenzyme of nucleotide nature, contains energy-rich (high-energy) phosphate bonds and can take part in the reactions of synthesis of complex substances, and also serve as a phosphate group donor.

Glutathione (γ-glutamyl-cysteinyl-glycine) is a peptide coenzyme.

Active center of the enzyme

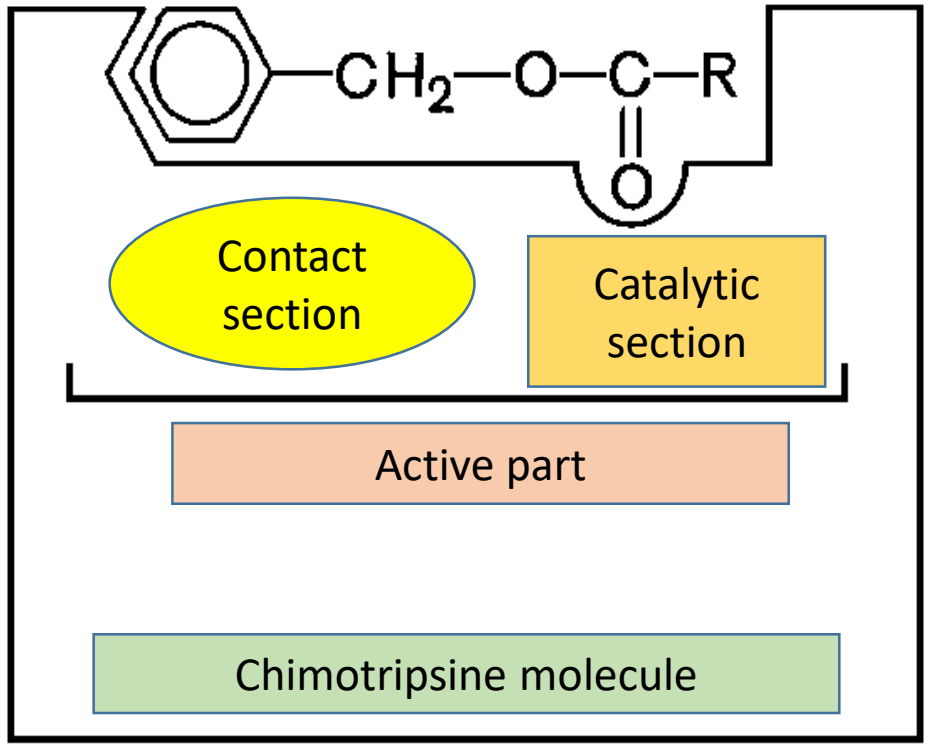
- A region of a protein molecule that can complementarily bind to a substrate and provide its chemical transformation. Side chains of amino acids and groupings of the non-protein part of the enzyme are involved in the formation of the active center.



Active center of the enzyme:

- occupies a relatively small part of the enzyme volume; the remaining amino acid residues provide the enzyme molecule with the correct globular shape;
- is formed at the level of the tertiary structure of the protein, groups belonging to different parts of the linear amino acid sequence participate in its formation;
- has the shape of a narrow depression or slot, into which water access is limited.

Active center of the enzyme:



Consists of two parts:

- **contact or anchor section** binding the substrate in the desired orientation;
- **catalytic section,** providing the reaction.

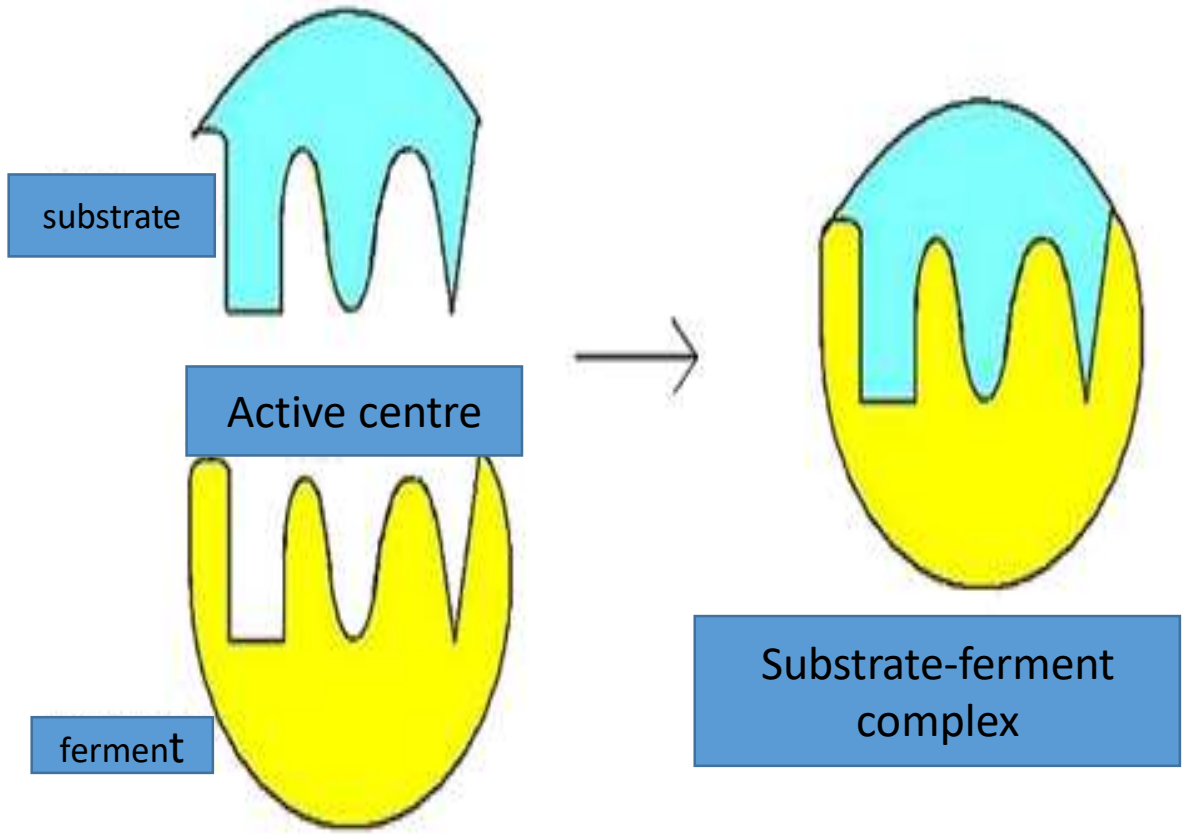
Interaction of the substrate with the active center

- Relatively weak interactions are involved in the formation of enzyme-substrate complexes:
- hydrogen bonds between polar uncharged groups of the substrate and the enzyme;
- ionic bonds between oppositely charged groups of the substrate and enzyme;
- hydrophobic interactions between non-polar groups of the substrate and enzyme.

Interaction of the substrate with the active center:

- model of strict correspondence ("key - lock"). The conformation of the active site is preliminarily adjusted to the shape of the substrate molecule.

- model of induced correspondence ("hand - glove"). The conformation of the active site is modified upon binding of the substrate.



Enzyme inhibition

- **Inhibition** - partial or complete inhibition of the enzymatic reaction under the influence of substances of various chemical nature.
- **Inhibitors** - substances that cause inhibition of enzymes.
- Depending on the **bond strength** enzyme inhibitor inhibition occurs **reversible and irreversible**.
- By **mechanism of action** the inhibitor is distinguished **competitive and non-competitive** inhibition.

Competitive inhibition

- the competitive inhibitor is structurally similar to the substrate;
- the competitive inhibitor interacts with the active center of the enzyme, preventing the interaction of the active center with the substrate;
- the effect of a competitive inhibitor depends on its concentration: the higher the concentration of the inhibitor, the lower the rate of the enzymatic reaction;
- the competitive inhibitor effect can be removed by increasing the substrate concentration.

Noncompetitive inhibition

- the non-competitive inhibitor is not structurally similar to the substrate;
- The noncompetitive inhibitor interacts not with the active site, but with other parts of the enzyme molecule. This leads to a change in the conformation of the active center and a decrease in the affinity for the substrate;
- the action of a non-competitive inhibitor does not depend on its concentration;
- the effect of a non-competitive inhibitor cannot be removed by increasing the concentration of the substrate;
- Non-competitive inhibitors decrease the molecular activity of the enzyme (turnover number).

Enzyme properties: oligodynamic

- High efficiency in very small quantities.
- For example, in the hydrogen peroxide splitting reaction:

$$2\ H_2O_2 \rightarrow 2\ H_2O + O_2$$

- One molecule of catalase at 0°C splits about 50,000 molecules of hydrogen peroxide per second.

Enzyme properties: specificity

- The selective ability of an enzyme to catalyze a well-defined reaction.
- The structure of the active site of the enzyme is complementary to the structure of its substrate. Therefore, the enzyme selects and attaches only its substrate from all the substances present in the cell.

Enzyme properties: specificity

- **Absolute specificity** - the selective ability of the enzyme to catalyze only one of the possible transformations of a single substrate.
    - **arginase** only catalyzes the hydrolysis of arginine,
    - **urease** only catalyzes the hydrolysis of urea.
- **Relative specificity** - the selective ability of the enzyme to catalyze the same type of transformation of substrates similar in structure.
    - **amylase** hydrolyzes glycosidic communication,
    - **pepsin and trypsin** - peptide bonds,
    - **lipase and phospholipase** - ester connections.
- **Stereochemical (optical) specificity** - the selective ability of the enzyme to catalyze the conversion of only one of the possible spatial isomers.
    - monosaccharide metabolism enzymes catalyze the conversion only D-isomers, but not L-isomers.
    - enzymes of amino acid metabolism in mammals, on the contrary, catalyzes the conversion of only L-isomers, but not D-isomers.

International classification and nomenclature of enzymes

- 6 classes of enzymes, depending on the type of catalyzed reaction;
- in each class several subclasses are distinguished;
- in each subclass - several sub-subclasses;
- in every sub-subclass - individual enzymes.
- Each enzyme is assigned an individual four-digit format code number:

Class_Number**...**Subclass_Number**...**Subclass_Number**...**Enzyme_Number_
_ in Subsubclass

The classification is based on the type of catalyzed reaction.

- Oxidoreductase catalyze oxidatively- restorative reactions.
- Transferases - group transfer reactions.
- Hydrolases - hydrolytic bond cleavage SS, FROMN, FROMS with water connection at the break point.
- Lyases - reactions non-hydrolytic splitting with the formation of double bonds, some reverse reactions synthesis.
- Isomerase - transfer of groups within the molecule with education isomers.
- Ligases catalyze the connection of two molecules, conjugate with a break pyrophosphate communication ATF.

Enzyme classification by source

Plant origin

-Dried milky papaya juice (proteolytic activity - caripazim);
-lekozyme - a mixture of three proteolytic enzymes: papain, chymopapain and lysozyme;
-nigedaza - enzyme with lipolytic activity, isolated from damask
- a complex of enzymes from bee pollen (pollen)

Animal origin

-pepsin (gastric mucosa)
-trypsin (pancreas)
-chymotrypsin (proteolytic activity
-RNase pancreatic
-DNase pancreatic
-collagenase

Microbiological enzymesical synthesis

- Streptoliasis from culture β- hemolytic group C streptococcus;
-Terrilitin - waste product Aspergillus terricola;
-Oraza - proteolytic enzyme from fungal culture Asp. Oryzae;
-RNase of Bacillus intermedicus

Classification of enzymes for medical purposes

With purulent-necrotic processes
Trypsin, Chymotrypsin
Terrilitin, RNase, DNase

Fibrinolytic

Fibrinolysin, Streptoliasis, Streptodecase, Celiasis

Enhancing digestion
- Pepsin, Pancreatin, Oraza, Nigedaza, Festal

Antitumor Asparaginase

Improving tissue permeability
Ronidase, Lidaza

Standardization of enzyme preparations

- In pharmacy, quality control of enzyme preparations is carried out by determining the units of catalytic activity contained in a unit mass of an enzyme preparation.
- The enzyme activity is judged by the amount of the converted substrate or the resulting reaction product for a certain period of time.
- Determination of activity is carried out under standard conditions: saturating substrate concentration, optimum pH of the medium, constant optimum temperature.
- By the decision of the International Biochemical Union, a standard unit of activity or International Unit of activity ME was adopted for the assessment of enzyme preparations
- ME is the amount of enzyme that, under given conditions, catalyzes the conversion of 1 micromole substrate in 1 min
- AU is a conventional unit, the value of which is indicated in private articles

Enzyme activity units

- ***Total enzyme activity*** - quantity micromole substrate, which undergoes transformation per unit of time based on the amount of biological material taken for research.
- Formula for calculation:

$$a = \frac{\Delta C}{B \times t} \times n,$$

where **a** is the enzyme activity (total), ΔC - the difference in substrate concentrations before and after incubation;

B is the amount of material taken for analysis, **t** is the incubation time; **n** - dilution.

•***Specific enzyme activity*** - quantity micromole substrate, which undergoes conversion per unit of time per 1 mg of protein in the test material. To calculate the specific activity of the enzyme, the total activity is divided by the protein content in the sample.

Standardization of enzyme preparations

- Enzyme preparations are standardized:

1.Specific activity of the drug - expressed in units of enzymatic activity of the enzyme (IU or U) per 1 mg of the drug and 1 mg of protein (the second indicator characterizes the purity of the drug)

2. The dose is expressed in units of enzymatic activity (IU or U) per unit of dosage form

- The conditions for the functioning of the gastrointestinal tract (pH, temperature, time) can be used as the conditions for the analysis. Such conditions make it possible to determine the activity with less accuracy, but the units obtained more characterize the behavior of the enzyme in the body, which also makes it possible to roughly estimate the effectiveness of the drug.
- Unit amylolytic activity - the amount of enzyme that, at a temperature of 30 ° C and pH 6.0 for 10 minutes, catalyzes hydrolysis to dextrins of various molecular weights 1 g of starch, which is 30% of the introduced into the reaction
- Unit lipolytic activity - the amount of enzyme, which at a temperature of 37 ° C for 1 hour releases 1 μmol fatty acid from 40% olive oil emulsion
- The unit of proteolytic activity is the amount of enzyme that, at a temperature of 37 ° C and pH 7.6 for 10 min, increases the optical density of the solution at a wavelength of 280 nm by 1.0.

Enzyme activity units

Using enzymes in medicine and pharmacy

- there are diseases caused by a congenital deficiency of certain enzymes in tissues (**enzymopathies**);
- determination of the activity of enzymes in blood and other tissues provides valuable information for diagnosis (**enzyme diagnostics**);
- enzymes or their inhibitors can be used as medicinal substances (**enzyme therapy**).

There are 4 directions to enzyme therapy:

1. Local use of enzyme preparations as nonspecific drugs, mainly for the removal of non-viable tissues (proteinases, depolymerase)

2. Parenteral use of enzymes for the purpose thrombolytic therapy, as well as the treatment of non-specific inflammatory diseases

3. Substitution therapy (replacement of the deficiency of digestive enzymes)

4. Use of enzyme inhibitors

Sources of obtaining enzyme preparations

1. Plant raw materials

1. A source of enzymes may be ***germinated grain of various cereals*** (malt):

- can either be used directly as *technical enzyme preparation*,

- *serve as source material* for obtaining purified enzyme preparations.

2. In tropical and subtropical countries as a raw material for industrial production ***proteinase*** use ***melon latex***, ***ficus latex*** (e.g. leaves, shoots *figs*, green mass juice *pineapple* and etc.)

Sources of obtaining enzyme preparations

***2. Organs and tissues of animals*.**

At all meat processing plants, raw materials containing enzymes are collected, preserved and used to obtain enzyme preparations.

So **raw materials are:**

- ***pancreas*** (contains chymotrypsin, collagenase, elastase, trypsin, amylase, lipase and dr),
- ***mucous membranes of pigs' stomachs and small intestines*** (pepsin and lipase),
- ***abomasum*** (pepsin and lipase),
- ***rennet of dairy calves and lambs*** (rennin (rennet extract)),
- testes of sexually mature animals (contain an enzyme hyaluronidase).

3. Microorganisms

Microorganisms can synthesize simultaneously a whole *enzyme complex*, but there are some, especially among the mutant strains, which are *monoenzyme* and form only one enzyme in large quantities.

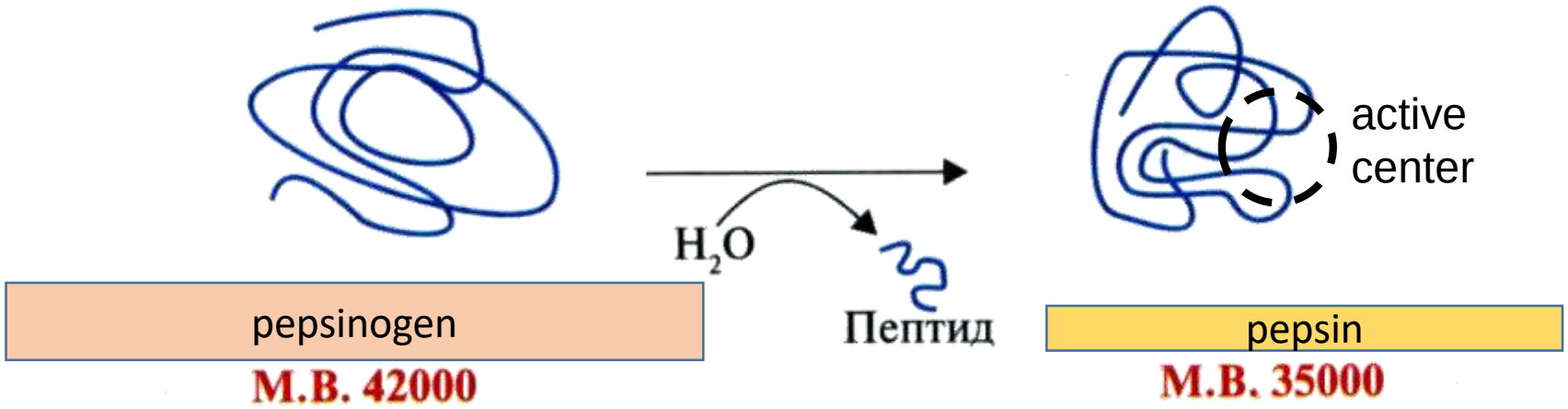
Benefits of microorganisms:

- *unpretentious to the composition of the nutrient medium*,
- *easily switch from the synthesis of one enzyme to another,*
- *have a relatively short growth cycle (16-100 hours).*

For industrial production of enzyme preparations, they are used as natural strains of microorganismsisolated from natural objects and mutant strains.

Producers enzymes can be: bacteria, mushrooms, yeast, actinomycetes.

Regulation by partial proteolysis



- As a result of hydrolysis of one or several peptide bonds, a part of the molecule is cleaved, and in the remaining part, a conformational rearrangement occurs and an active center is formed.

Getting pepsin

- Grinding raw materials (tops mills)
- Extraction with wateracidified with acid hydrochloric to the value

pH 1.9-2.3 (method bismaceration, the ratio raw materials and extractant 10: 1 extractio

8 h again 24 h, extraction temperature 40 ° C).

- Cleaning lysates (separated from the top layer of fat, filtered. pH lysate 1.9-2.3)
- Union lysates
- Isolation of pepsin (method salting out - to lysate at continuous stirring add 20-25% sodium solution chloride. The released pepsin floats to the surface).
- Filtration (pepsin is separated)
- Drying (dried in a vacuum drying oven at a temperature 35-40 ° C)
- Grinding (grind in a porcelain ball mill and sift)
- Standardization (by proteolytic activity -digestion chicken protein eggs: 10 g of mashed protein in the presence of 0.1 g of the drug under standard conditions should completely dissolve in 3-4 hours, forming an opalescent solution. After determining the biological activity, the drug is mixed with powdered sugar.

Pepsin is a slightly yellowish powder, sweet taste with a weak peculiar smell.

Biotechnological production of enzyme preparations

The feasibility of using microorganisms is due to:

one) *genetic manipulation* it is possible to increase the level a thousand or more times *catabolic* and several hundred times the level *biosynthetic enzymes*;

2) *energetically justified* growing microbial cells on a large scale in connection with the use inexpensive media and rapid growth of microorganisms;

3) *a huge variety of reactions*that microorganisms are capable of (especially with regard to secondary metabolism);

4) microorganisms *serve as a source of a number of unique enzymes*not found anywhere else (**cellulose, tannase, hydrogenase**, **keratinases, nitrogenase, penicillinase and etc**.)... there is species developing at extremely high temperatures (87 ºC), in connection with which it is potentially possible **creation of thermostable strains**;

✖ 5) *ability to adapt to different conditions*, which allows you to transfer the culture to production, cultivation on cheap substrates.

Industrial **production and use of enzymes** based on two important factors:

- At first, *enzymes are produced in living cells*;

- Secondly, *enzymes can exert their effect in the environment independently of living cells*.

Requirements for the producer:

1) education is desirable **extracellular enzymes**since they are easier to highlight;

2) **high enzyme yield** for a short time;

3) **enzyme purification** from cultural liquids **should be easy**;

4) **strains should not produce antibiotics**, toxic substances and should not be related to strains that generate toxins.

Cultivation technology of producers

✖ The technological process can be broken down into three stages:

one) *obtaining seed*;

2) *obtaining a production culture* methods superficial or deep cultivation;

3) *excretion* from a finished production culture technical or purified *enzyme preparations*.

Surface method consists in the cultivation of microorganisms on the surface of moistened sterilized bran placed in cuvettes (incubation is carried out in a special thermostatically controlled workshop with constant control of temperature, humidity and air supply in it).

Deep methodmore economical. For its implementation, stainless steel fermenters are used, equipped with devices for mixing and feeding sterile air into the liquid nutrient medium.

Getting active producers.

1. Active strains identified from natural sources, *exposed to mutagens multiple times*, i.e. **carry out stepwise selection**. The combined effect of chemical and physical mutagens is often effective.

So, application **ethyleneimine** and **ultraviolet radiation** in *combined with step sampling* allowed to obtain very active strains ***Asp. Awamori*** used as producers ***amylolytic, proteolytic and other enzyme complexes***. Selectionproduction valuable strains are also carried out under production conditions.

2. Activities for **preservation of producers**.

There are a number of storage methods productionvaluable strains providing their high biochemical activity. In museums of living cultures at factory laboratories, cultures are periodically sown and after the culture has developed in a dense medium, it is poured with sterile vaseline oil. In many cases, lyophilization of cultures is the best storage method.

Culture media for the cultivation of microorganisms...

By appointment divided into groups:

1. Wednesday to maintain producer strains in the laboratory and in the museum (selected based on the physiological needs of microorganisms).
2. Wednesday to obtain seed.
3. High volume media in the main production process.

Structure media providing the accumulation of one or another enzyme may differ significantly from the composition of media used to isolate and maintain the culture of producers.

Some enzymes of microorganisms require the presence in the environment for their synthesis.

Inductor. So, for example, if for synthesis **lipases** *Aps. niger* favorable presence in the environment vegetable oil, and for synthesis **ribonuclease** - nucleic acids, then for increased synthesis **α-amylase** *You. polymyxa* need to add hydrolyzate casein.

Technological features of fermentation processes

By technological design distinguish between processes:

- *aerobic* cultivation,
- *anaerobic* cultivation;
- *solid phase* cultivation,
- *superficial* cultivation,
- *deep* cultivation:
  - *periodic* cultivation
  - *continuous* cultivation.

Dosage forms of enzymes

- The choice of Lf enzymes is due to their physicochemical properties, therapeutic activity and degree of purification. For example, for enzyme preparations used to treat purulentnectrotic processes, the most acceptable form of release is a powder, which is diluted immediately before use in a sterile solvent and is used in the form of injections, irrigations, inhalations (trypsin, recommended for the removal of purulent exudates upper respiratory tract). Himopsin as a less purified preparation is recommended only locally in the form of applications and inhalations.
- The widest range of choice of LF in FP that improve digestion processes (pepsin - powder for the manufacture of medicine, pancreatin - enteric coated tablets, oraza- granules). Complex enzyme preparations have a complex structure.

Dosage forms of enzymes

MEXAZA

A drug released in the form of a three-layer dragee:

- **inside is 0.1 g enteroseptol and 0.01 g entobexa** (antibacterial effect, suppress the vital activity of protozoa, including amoebas)
- **the second layer contains 0.15 g of pancreatin and 0.025 g dehydrocholic acid** (pancreatin is a digestive enzyme that replenishes the deficiency of pancreatic enzymes; dehydrocholic acid stimulates the pancreas, helps in the production of bile and emulsifies fats
- **outside - 0.05 g of the enzyme component bromelain** (bromelin - a mixture of plant-derived enzymes that promote protein processing. It acts in the stomach and intestines, stimulates digestion, cleanses the blood, strengthens the immune system.Bromelin helps to improve the breakdown of food, in particular, protein, in a fairly wide range of gastric acidity - from pH = 3.0 to pH = 8.0. The maximum possible is pH = 0.86, and the minimum is pH = 8.3.Bromelin effective for any gastric acidity juice.

Dosage forms of enzymes

MICRASIM - pancreatin pellets in capsules

- **The form release, composition and packaging**

◊ ***Capsules*** hard gelatinous, from transparent body and painted lid. Cap color depends on drug dosage and capsule size;

content capsules - enteric pellets cylindrical or spherical, or irregular in shape from light brown to brown, with a characteristic odor; color variation allowed...

- IN the composition of the drug includes natural enzymes from the pancreas of animals - ***protease, lipase and amylase***, providing the digestion of proteins, fats and carbohydrates from food.
- After taking the drug Micrasimthe capsule dissolves rapidly in the stomach, releasing enteric-coated pancreatin pellets. Due to their small size, the pellets are quickly and evenly mixed with food and, simultaneously with the food lump, easily penetrate into the duodenum, and then into the small intestine, where pancreatic enzymes are released and begin to actively act, contributing to the rapid and complete digestion of proteins, fats and carbohydrates from food.
- Fast stirring pellet pancreatin with the contents of the stomach, their uniform distribution in it, simultaneous passage with chyme, as well as the preservation of enzymes before they start to work in the intestine (due to the presence of an enteric membrane pellet), provide a higher digestive activity and the maximum approximation of the action of the drug to the natural process of digestion.

Dosage forms of enzymes

Creon

- Digestive enzyme agent.
- Release form: capsules enteric, gelatinous, solid, with a brown opaque lid and a colorless transparent body; content- minimicrospheres light brown (20, 50 and 100 pcs. in plastic bottles with a screw cap and first opening control, 1 bottle in a cardboard box).
- Active substance of the preparation: pancreatin, in 1 capsule - 150 mg, which corresponds to the content: lipase - 10,000 units of measurement of the European Pharmacopoeia (ED Eur. F.); amylase - 8000 UNITS Eur. F .; protease - 600 units Eur.F.
- Additional components (their content in 1 capsule): excipients: hypromellose phthalate (56.34 mg), macrogol 4000 (37.5 mg), triethyl citrate (3.13 mg), dimethicone 1000 (1.35 mg), cetylalcohol (1.18 mg); composition of the capsule shell: gelatin (60.44 mg), sodiumlauryl sulfate (0.12 mg), titanium dioxide (E 171) (0.07 mg), as well as dyes of iron oxide yellow (E 172) (0.05 mg), iron oxide red (E 172) (0.23 mg) and iron oxide black (E 172) (0.09 mg).

Dosage forms of enzymes

PANCITRATE

- Digestive enzyme agent.
- **Release form**: Elongated gelatin capsules containing microtablets with enteric coating in glass / polyethylene bottles in carton packs # 20, 50, 100.
- **Ingredient: Pancreatin**, crospovidone, microcrystalline cellulose, aerosil, stearate magnesium, triethyl citrate, copolymer ethyl acrylate and methacrylic acid, talc, emulsion simethicone, gelatin, glycol Montan wax, titanium dioxide, iron oxide red / black.
- A drug compensates for the function of the pancreas caused by insufficient secretion production. Possesseslipolytic, proteolytic and amylolyticaction. Under the influence of enzymes**pancreatin** the process of splitting proteins occurs to amino acids, starch - up to **monosaccharides** and **dextrins**, fats - to fatty acids and glycerin. Pancitrate normalizes all digestive processes, improves function **Gastrointestinal tract**... From the dosage form, pancreatic enzymes are released in the small intestine in an alkaline environment; capsules are protected from the action of gastric juice by a resistant membrane.
- After getting into the stomach, the contents of the capsule of the drug are mixed with the food taken and distributed evenly in the acidic environment of the stomach. Acid-resistant coating when passed through the stomachmicrotablets protects enzymes **pancreatin** from inactivation... With further advancement with food into the small intestine, a coating that is resistant to stomach juicemicrotablets dissolves in an alkaline environment and enzymes are quickly released...

Technological scheme for obtaining tabletsSomilaza" (contains lipolytic drug from Penicillium solitum and α-amylase

TP-1. **Granulate production solizima** - substance solizimairrigated with PVP solution in chloroform. The wet mass is dried in a fluidized bed in a stream of nitrogen

TP-2. **Granulate production α-amylase -** substance α-amylase irrigated with PVP solution in chloroform. The wet mass is dried in a fluidized bed in a stream of nitrogen

TP-3. **Getting a tablet mass -**The mixer is loaded granulate solizima and α-amylases, sugar granules, stearic acid. Mix thoroughly.

TP-4. **Pressing** - Core tablets are produced on RTM at low pressing pressures using a spherical press tool.

TP-5 **Film coating** - a solution of APC, plasticized with vaseline oil in a fluidized bed in a stream of nitrogen, is applied to the core tablets.

TP-6. **Packing and packaging** enteric-coated tablets, 10 pieces each, are packed in a blister strip of PVC film and aluminum foil.

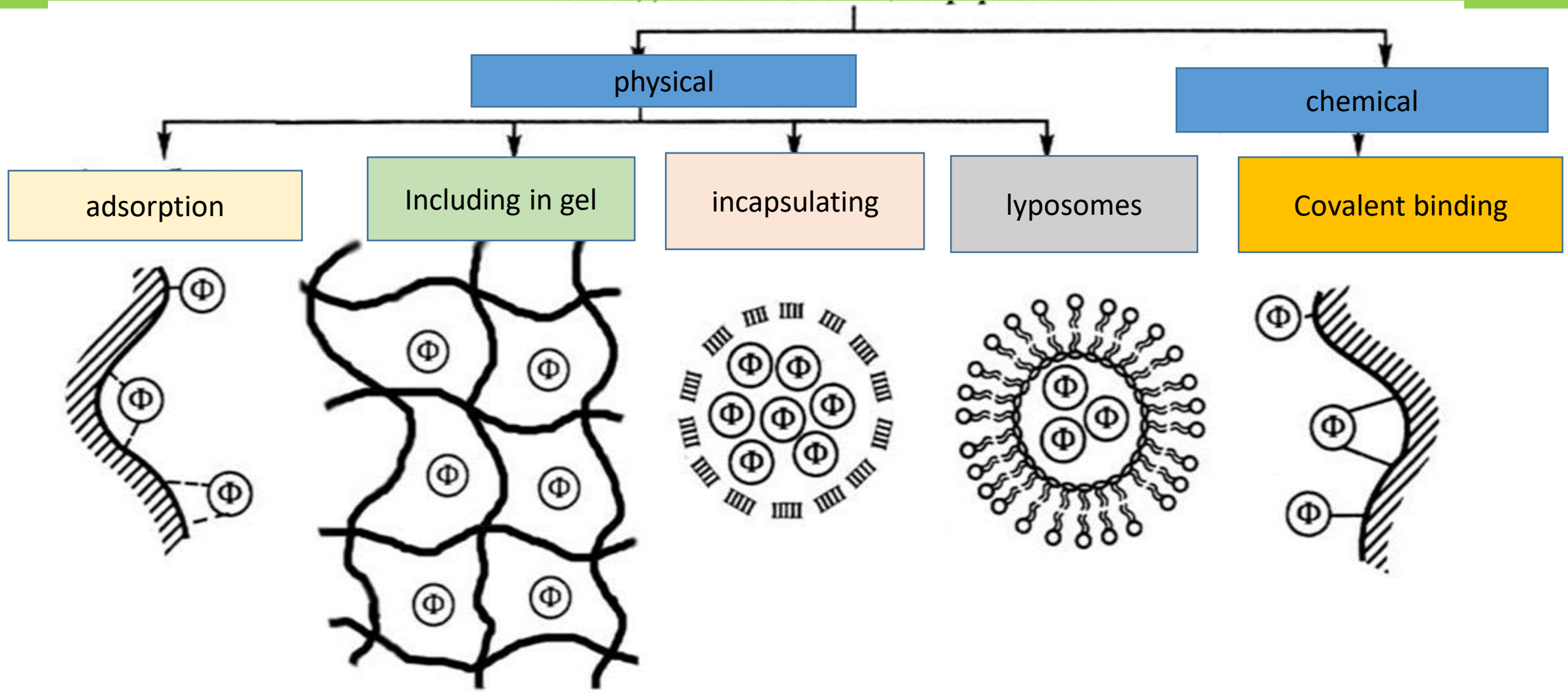
Enzyme immobilization

- Immobilization (lat. immobilis - immobile) - binding of enzyme molecules to the carrier
- Advantages: resistance to heat, changes in the pH of the medium, insolubility in water, facilitates separating them from the reaction products, provides a prolonged action and the possibility of repeated use
- Carrier: polymer (cellulose, polyacrylamide, agarose and etc.)
- Immobilization methods:

 - physical

 - chemical

Enzyme immobilization methods

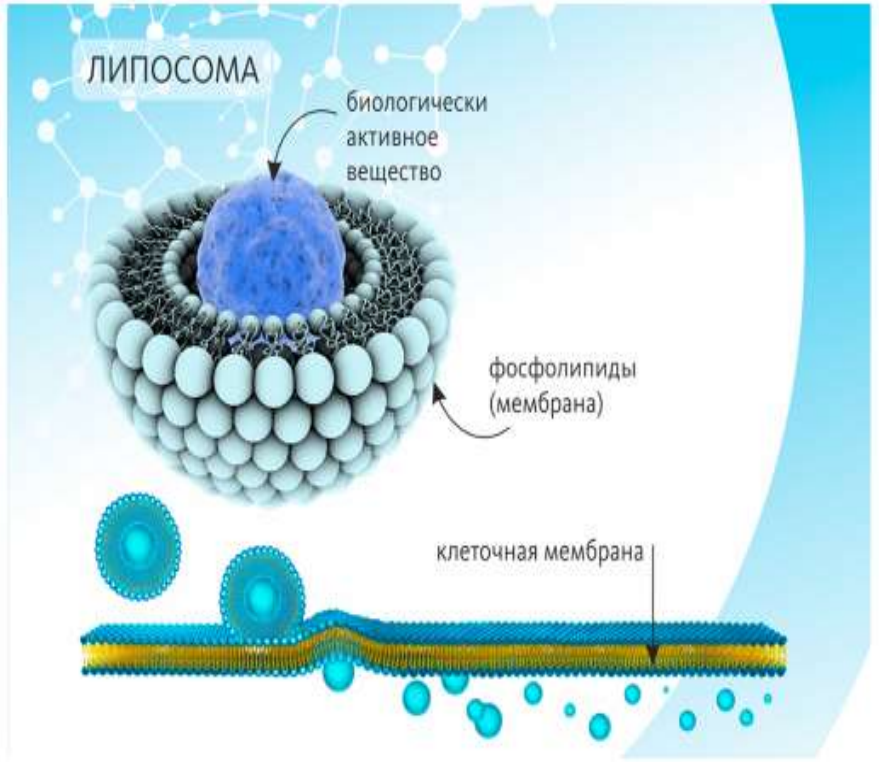


Φ - enzyme

Immobilization enzymes - physical methods

fixing the enzyme on a polymer carrier (polystyrene)

Incorporation of an enzyme into liposomes



Chemical methods of enzyme immobilization

- Based on the formation of new covalent bonds between the enzyme and the carrier.
- Unlike physical methods, these methods provide strong and irreversible bond of the enzyme with the carrier and are accompanied by the stabilization of the enzyme molecule.