

Enzymes (1. LF VL, Medical chemistry and biochemistry 1. parallel)

Enzymes are proteins with biocatalytic function. They are a tool of expression genes. The result of their action in the organism is a substance transformation (intermediate metabolism), an expression of the balance between anabolic and catabolic processes.

In catabolic, the organism gains energy for mechanical or osmotic work, but also for anabolic (e.g. synthesis of other precursors, macromolecular substances, mediators and hormones, necessary for vital functions and their regulation). Disadvantageous metabolic steps are overcome by coupled enzyme reactions (connection of reactions endergonic with exergonic).

- Substances that are changed by the action of enzymes are called substrates.
- The main features of biocatalysis are particularly significant catalytic efficiency and greater or lesser specificity to the type of substrate and to the type of catalyzed reaction.
- The protein nature of the enzyme and its specific interaction with the substrate resulting in the formation of an enzyme-substrate complex also result in other properties of enzyme systems, such as the influence of factors of a physical and chemical nature. The following influence here:
 - temperature
 - pH of the environment,
 - ions as activators, etc.
 - inhibitors (competitive or non)
 - effectors allosteric nature

Division of enzymes

Enzymes are divided into seven main classes according to the type of reaction they catalyze.

1. Oxidoreductases - dehydrogenases, oxidases, peroxidases, oxygenases
2. Transferases - aminotransferases, kinases
3. Hydrolases - peptidases, glycosidases, phosphatases
4. Lyases - synthases, decarboxylases
5. Isomerases
6. Ligases
7. Translocases

Usage of affinity chromatography

The specific interaction of an enzyme with a substrate, substrate analogue or inhibitor can also be used during enzyme isolation. However, the enzyme as a protein can be isolated by procedures common in protein biochemistry. Recently, it has been used with advantage affinity chromatography.

- This method uses the specificities of biological interactions such as: enzyme-substrate, enzyme-substrate analogues or enzyme-inhibitor.
- There are other options for specific interactions, such as type antigen-antibody, hormone carrier (receptor), etc.

Affinity chromatography is a special case of adsorption chromatography, in which the adsorbent has a specific affinity for the substances to be isolated.

- During enzyme isolation, an inhibitor or substrate analogue is bound to a suitable carrier (e.g. Sepharose[®], which is agarose gel prepared in the form of spherical particles). Such a bound group with a specific affinity is called ligand (affinant).
- It is often necessary to distance the affinant from its own carrier by means of a so-called spacers that steric hindrances do not occur when a large molecule of the enzyme (protein) approaches a small molecule of the bound ligand.
- The binding of the enzyme to the ligand is reversible, and this reversible binding can be disrupted by changing the physico-chemical properties of the washing fluid, e.g. by changing the pH, ionic strength, or by an excess of free substrate or substrate analogue, which displaces the ligand from binding to the enzyme.

Mechanism of action of enzymes

Enzymes work on the principle of lowering the activation energy. An enzyme-substrate (ES) complex is formed, then an enzyme-product (EP) complex is formed, which disintegrates to release the product.

Iron

Physico-chemical influences affecting the activity of enzymes

__ Physico-chemical influences affecting the activity of enzymes

Determination of enzyme activity

__ Determination of enzyme activity

References

References

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