Modulation of the activity of an already existing enzyme

Covalent modification of enzyme molecule

Formation of active enzymes from inactive precursors

Many enzymes are formed in an inactive form (so-called **proenzymes *or*** *zymogens*). Partial proteolysis of the enzyme changes the molecule into an active form and increases the concentration of the active enzyme. Eliminating the enzymes activated in this way arranges their proteolytic breakdown. This process is typical, for example, for digestive enzymes or for some factors coagulation cascades.

Enzyme Interconversion

Enzyme interconversion is the rapid switching between the active and inactive form of an enzyme molecule by another enzyme. The best-known example is the reversible ATP-dependent phosphorylation and dephosphorylation of the hydroxyl groups of the amino acids serineu, threonineu or tyrosineu forming the chains of the enzyme.

Some enzymes are ``activated *by phosphorylation (eg glycogen phosphorylase), others are ``inhibited* by phosphorylation (eg glycogen synthase). Phosphorylation is catalyzed by enzymes belonging to ``protein kinases *(phosphotransferases), dephosphorylation is provided by ``protein phosphatases.* , for example, in response to a hormonal signal (the connection between the signal coming to the cells from the outside and subsequently influencing the events inside them).

Interferences that affect enzyme kinetics

Effect of substrate and product concentration, K_M value, pH, temperature, etc.

- **Availability of substrates**

  The concentration of regulatory enzymes of metabolic pathways is very low in the cell. Likewise, the concentration of substrates is much lower than the value of the Michaelis constant (K_M coincides with the substrate concentration at which the rate of the enzyme-catalyzed reaction reaches half the maximum rate). Even a slight change in the concentration of the substrate will change the speed of its transformation.

  > *The change in substrate concentration takes place through increased intake, synthesis or transport of the substrate to the place where we metabolize it.*

- **Effect of the Michaelis constant K_M**

  Enzymes have a certain substrate specificity. If an enzyme converts several different substrates, it has a different affinity for each substrate. If, for example, two different enzymes can convert the same substrate, each of them has a different affinity for the given substrate. The higher the affinity to the substrate (the lower the K_M for the enzyme-substrate pair), the lower the concentration of the substrate around its active center is sufficient for the enzyme, which is necessary for the reaction to take place.

- **Removing Products**

  If the product of the reaction is used immediately, it does not accumulate and the reaction continues in the direction of its further formation. If the unused product begins to accumulate, it often serves as an inhibitor of the reaction or sequence of reactions leading to its formation. The following mechanisms prevent this in metabolism:

  1. removal of the product of one reaction by a subsequent reaction (principle of metabolic pathways),
  2. utilization of the product of one metabolic pathway in another metabolic pathway,
  3. product transport to another cellular compartment.

  All these processes will speed up the course of the given reaction.

- **Effect of pH**

  A change in pH can also affect enzyme activity by changing the dissociation of functional groups in the enzyme's *active center* (electrostatic interactions during substrate binding) and in the enzyme's *whole molecule* (change in the biologically active conformation of the enzyme in a less active conformation – e.g. access to the active center).

Effect of the presence of activity modulators (activators or inhibitors)

The activity of a regulatory enzyme can be affected by the direct binding of some substance (ligand or effector or modulator) to its protein molecule. A positive effector serves as an activator of the enzyme, while a negative effector is its inhibitor.

- Accumulation of the final product (or intermediate) of a given metabolic pathway often leads to the inhibition of the regulatory enzyme of the given pathway, to which the product binds - the so-called '*regulation from fifth bond*.
- The (inter)product of one metabolic pathway can influence (activate or inhibit) the activity of the regulatory enzyme of another, somehow related metabolic pathway – the so-called *cross-regulation*.
- An intermediate product of a metabolic pathway can influence the activity of a subsequent enzyme of a given metabolic pathway – the so-called '*step forward regulation*.

🔎 *For more information see Enzymes (FBLT).*

Modulators

Modulators' can bind to the enzyme either directly in the active center (competitive inhibition), or they bind to another, so-called **allosteric site** (allosteric regulation). Natural activity modulators bind to the enzyme non-covalently, using only weak non-bonding interactions.

Isosteric modulation of enzyme activity

Isosteric modulation of enzyme activity refers to **simple enzymes** that exhibit a hyperbolic dependence between reaction rate and substrate concentration. Their activity is mainly affected by changes in substrate concentration, reduction or increase in enzyme synthesis. In addition, enzyme activity is affected by inhibitors that bind directly to the active center instead of the substrate (competitive inhibition). A competitive inhibitor can be one of the products of the metabolic pathway (feedback inhibition).

Allosteric regulation

Allosteric regulation occurs in **multisubunit enzymes'** (most regulatory enzymes of metabolic pathways). These enzymes show a ``sigmoid dependence *of reaction rate on substrate concentration. Allosteric modulators of activity bind outside the **active center** to other sites on the enzyme molecule. The binding of the modulator changes the conformation of the molecule, which can be manifested by a* change in the affinity *of the enzyme to the substrate (ie, a decrease or increase in the K_M value).*

The concentration of the active enzyme can also be reduced (part of the enzyme molecules is inactivated), thereby causing a change in the value of the maximum rate of the enzyme-catalyzed reaction. Due to the binding of the activator, the less active so-called "T-form" of the enzyme ("tension") changes to the more active "R-form" ("relaxed"). The binding of allosteric activators can thus increase the number of enzyme molecules in the R-form.

'*Substrates and effectors*' ***generally only affect the balance between the T and R conformations - the two conformations coexist in different proportions. The final amount of active forms of the enzyme depends on the total effect of the various activators and inhibitors binding to the enzyme, i.e. it depends on their*** *mutual ratio* in the cell. With an increase in K_M, under physiological conditions, the reaction can be completely eliminated, since the physiological concentration of the substrate lies in the region where the enzyme is practically ineffective. It is a very fine and continuous regulation of the reaction rate based on the connection of various metabolic pathways.

- ws: Modulace aktivity již existujícího enzymu