Enzyme

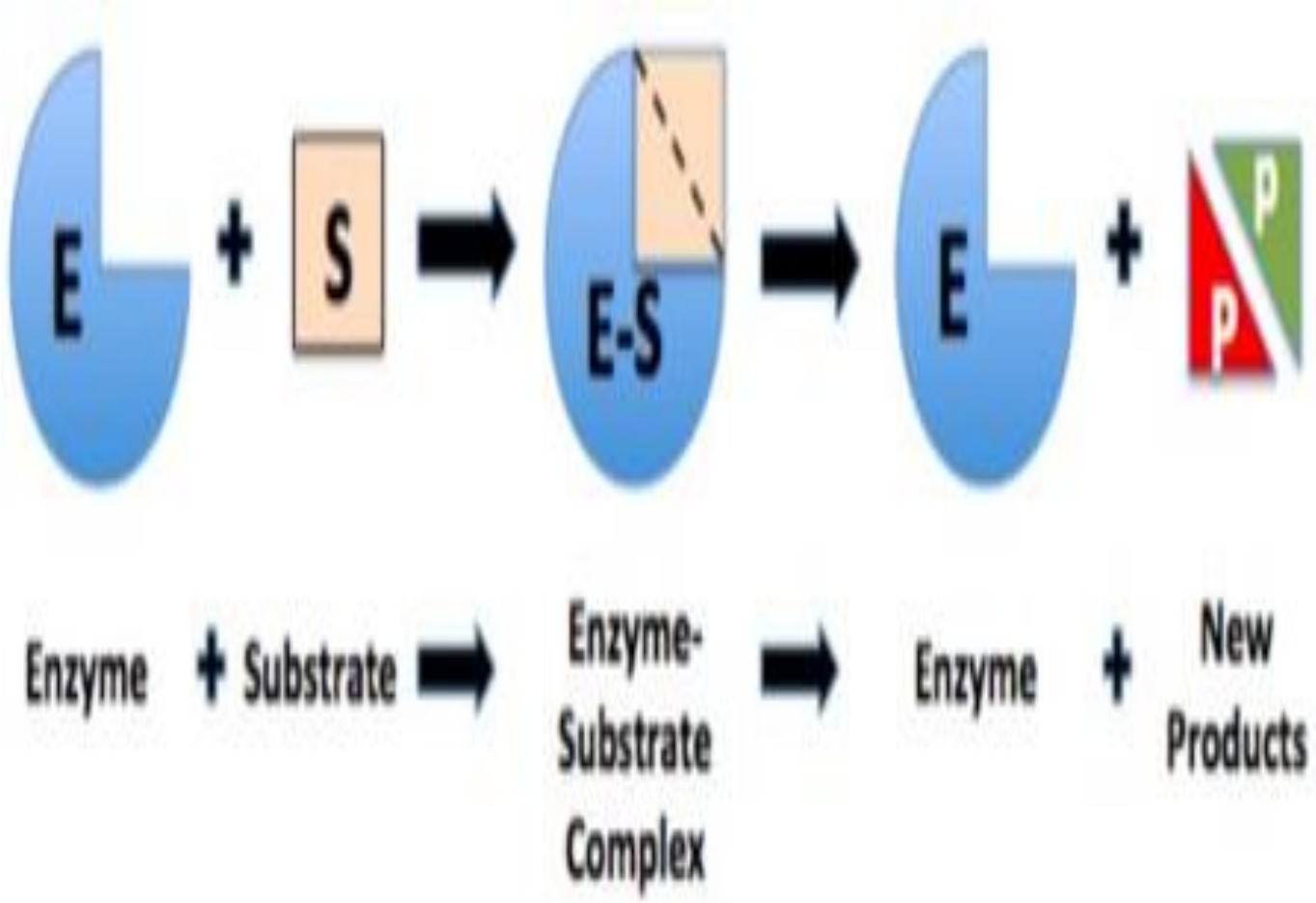
Coenzyme

Apoenzyme

Holoenzyme

Cofactor

Enzymes temporarily combine with the chemicals involved in a reaction. These chemicals are called the **substrate**. The combination is called the enzyme-substrate complex. When the enzyme and substrate combine, the substrate is changed to a different chemical called the product. **The enzyme is not consumed or altered by the reaction.**



Substrate Concentration

It has been shown experimentally that if the amount of the **enzyme is kept constant** and the **substrate** concentration is then **gradually increased,** the reaction **velocity will increase** until it reaches **a maximum**.
After this point, increases in substrate concentration will not increase the velocity.

Four Steps of Enzyme Action

1-The enzyme and the substrate are in the same area.

2. The enzyme grabs on to the substrate at a special area called the active site.

The combination is called the **enzyme/substrate complex**. Enzymes are very, very specific and don't just grab on to any molecule. The active site is a specially shaped area of the enzyme that fits around the substrate. It can only pick up one or two parts.

3. A process called catalysis happens.

Catalysis is when the **substrate is changed**. It could be broken down or combined with another molecule to make something new. It will break or build chemical bonds. When done, it will have the **enzyme/products complex**.

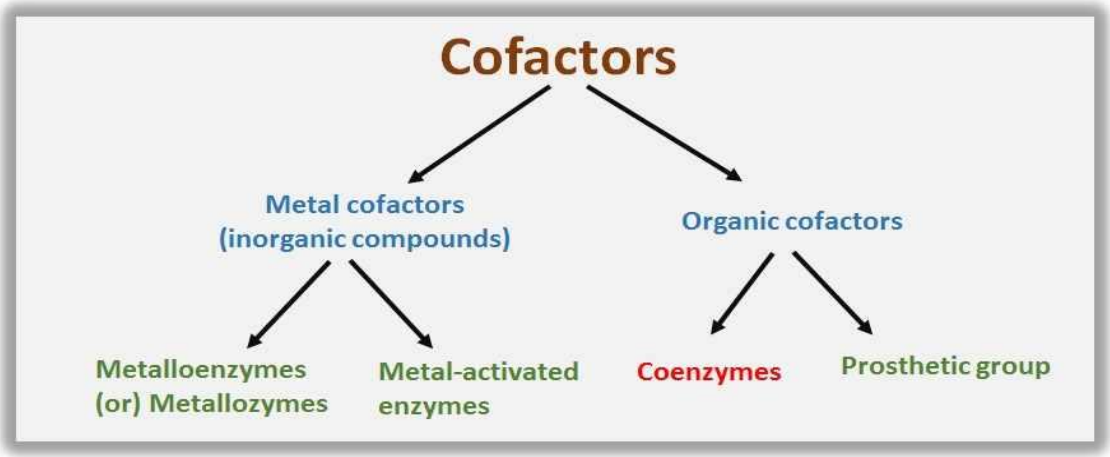
4. The enzyme releases the product. When the enzyme lets go, it returns to its original shape.

It is then ready to work on another molecule of substrate.

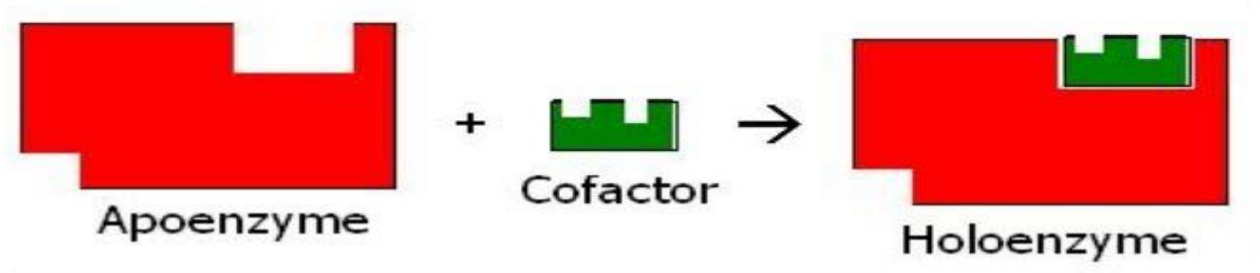
Cofactor

A cofactor is the **non-protein part** of an enzyme that is essential for the enzyme's activity as a catalyst.

Cofactors can be differentiated into organic and inorganic.

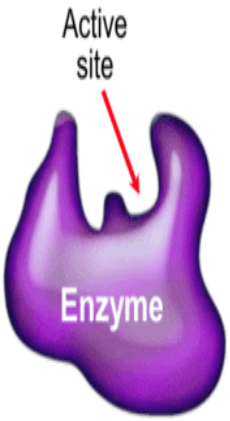


Together with the **apoenzyme (protein component),** form the complete **enzyme (holoenzyme**). The removal of the cofactor from an enzyme results in the loss of enzymatic activity.

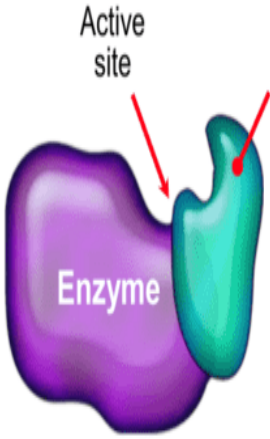


Enzyme Cofactors

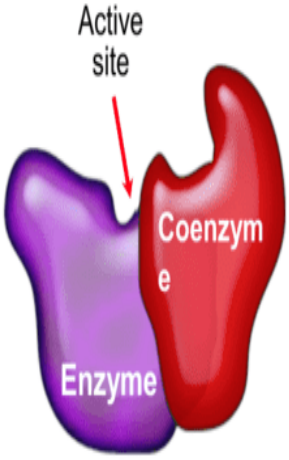
- Some enzymes require **cofactors** to be active.
- Cofactors are a **nonprotein** component of an enzyme. Cofactors can be:
  - **organic molecules** (coenzymes).
  - **inorganic ions** (e.g. Ca^{2+}, Zn^{2+}).
- Cofactors may be:
  - **Permanently attached**, in which case they are called **prosthetic** groups.
  - **Temporarily attached coenzymes**, which detach after a reaction, and may participate with another enzyme in other reactions.



Enzyme is protein only
Example: lysozyme



Enzyme + prosthetic group
Example:
flavoprotein + FAD



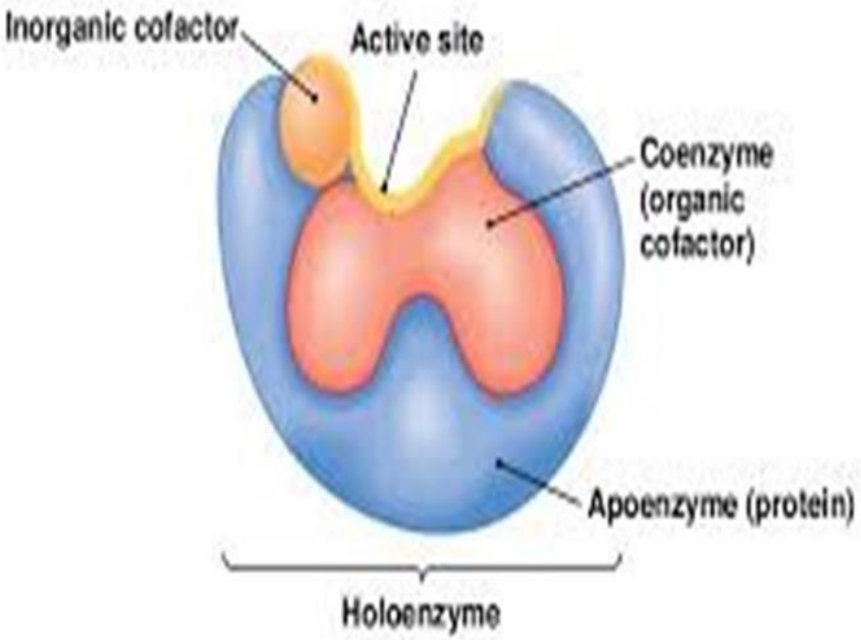
Enzyme + coenzyme
Example:
dehydrogenases + NAD

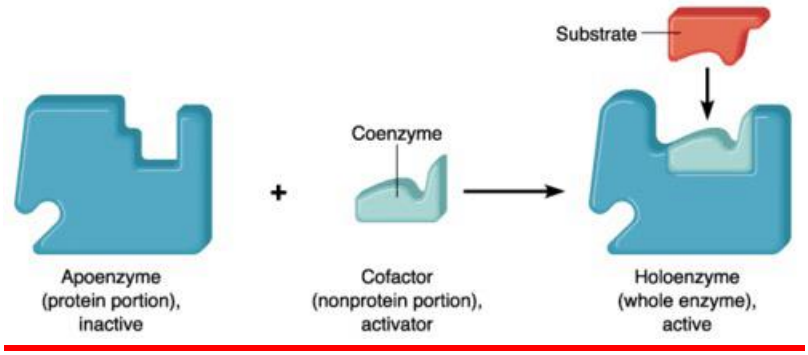
Cofactor vs Coenzyme

A cofactor is a non-protein chemical compound.	**A coenzyme is a specific type of cofactor that is organic.**
Cofactors can be inorganic or organic molecules.	**Coenzymes are exclusively organic molecules.**
Inorganic cofactors include metal ions like Mg^{2+} or Fe^{2+}.	**Coenzymes often include vitamins or derivatives like NAD^{+}.**
Cofactors bind to enzymes to assist in catalysis.	**Coenzymes bind loosely to enzymes and are often carriers of specific atoms or functional groups.**
Cofactors are not consumed in the reaction.	**Coenzymes are typically altered during the reaction.**
Cofactors can be permanently bound to the enzyme.	**Coenzymes are usually transiently bound and may need to be regenerated.**
Cofactors can stabilize enzyme structure.	**Coenzymes often serve as intermediate carriers of electrons or chemical groups.**
The role of a cofactor is often structural or catalytic.	**The role of a coenzyme is to transfer chemical groups from one substrate to another.**
Some cofactors are essential for enzyme activity.	**Coenzymes often act as a secondary substrate in enzymatic reactions.**
Cofactors may include both organic and inorganic molecules.	**Coenzymes are exclusively organic and are usually derived from vitamins.**

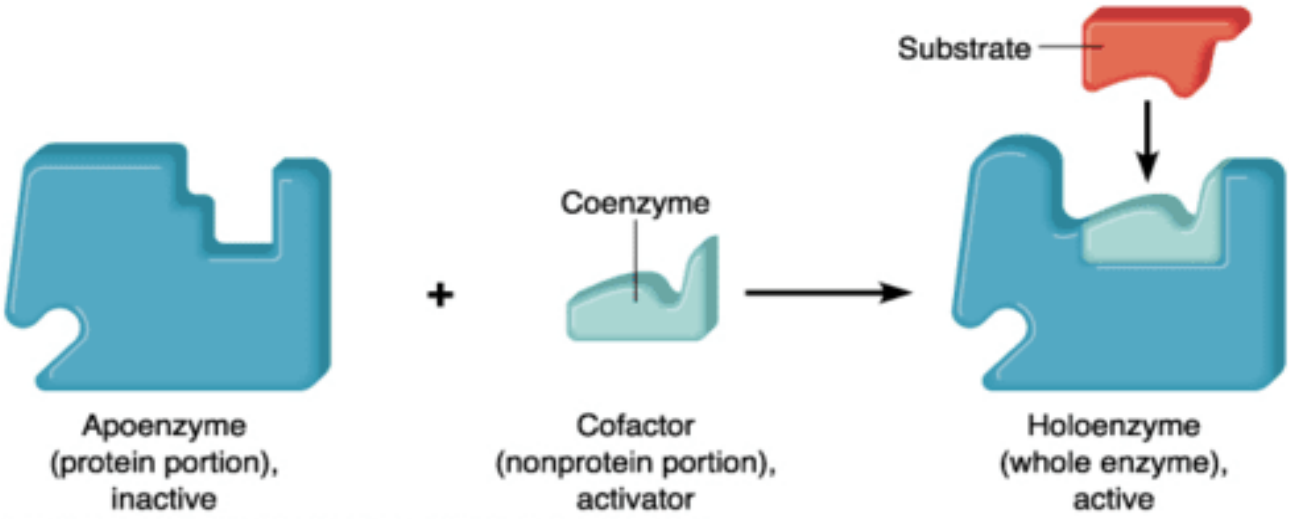
Holoenzymes

- Some enzymes require molecules other than proteins for enzymatic activity.
- The term **holoenzyme** refers to the **active** enzyme with its **nonprotein** component.
- The term **apoenzyme** is **inactive** enzyme without its **nonprotein** part.
- If the **nonprotein part** is a **metal ion** such as Zn 2+ or Fe2+, it is called a **cofactor**.
- If it is a **small organic molecule**, it is termed a **coenzyme.**





Apoenzyme is the protein part of an enzyme , are important for enzymatic activity since they are responsible for the specificity of enzymes to their substrates. Apoenzymes alone are not active enzymes; they **must bind** to an organic or inorganic cofactor in order to be activated.



Apoenzyme + Cofactor = Holoenzyme

The cofactor may be:

1. A coenzyme - a non-protein organic substance which is dialyzable, thermostable and **loosely attached** to the protein part

2. A prosthetic group - an organic substance which is dialyzable and thermostable which is **firmly attached** to the protein or apoenzyme portion.

3. A metal-ion-activator - these include K+, Fe++, Fe+++, Cu++ ,Co++ , Zn++ , Mg++ ,Ca++ , and Mo+++

Coenzymes

Coenzyme are a small, organic, non-protein molecules that carry chemical groups between enzymes.. These molecules are called **coenzymes** because they work together with enzymes to enhance reaction rates.

In contrast to substrates, coenzymes are not irreversibly altered by the reactions in which they are involved. Rather, **they are recycled** and and can participate in multiple enzymatic reactions.

Prosthetic groups are small molecules bound to proteins in which they play critical functional roles.

Coenzymes serve as carriers of several types of chemical groups. An example of a coenzyme is **nicotinamide adenine dinucleotide** (**NAD^+**), which functions as a **carrier of electrons in oxidation-reduction reactions .**

NAD^+ can accept a hydrogen ion (H^+) and two electrons (e^-) from one substrate forming NADH, then NADH can donate these electrons to a second substrate, re-forming NAD^+.

Thus, NAD^+ transfers electrons from the first substrate (which becomes oxidized) to the second (which becomes reduced).

Many coenzymes are closely related to vitamins, which contribute part or all of the structure of the coenzyme. Vitamins are necessary components of the diets of human.

Examples of Coenzymes and Vitamins

Coenzyme	Related vitamins	Chemical reaction
NAD^+, $NADP^+$	Niacin (B_3)	Oxidation-reduction
FAD	Riboflavin (B_2)	Oxidation-reduction
Thiamine pyrophosphate	Thiamine (B_1)	Aldehyde group transfer
Coenzyme A	Pantothenate(B_5)	Acyl group transfer
Tetrahydrofolate	Folate (B_9)	Transfer of one-carbon groups
Biotin	Biotin(B_7)	Carboxylation
Pyridoxal phosphate	Pyridoxal (B_3)	Transamination

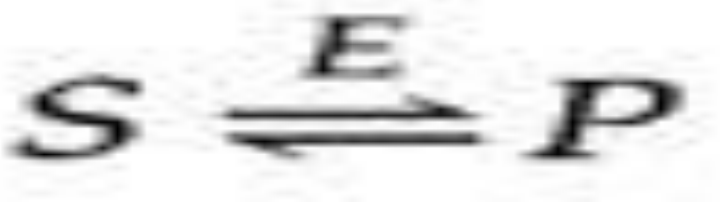
The Catalytic Activity of Enzymes

An enzyme acts by **lowering the activation energy** of a chemical reaction, which increases the rate of reaction. Activation energy is defined **(The energy required to convert all molecules of a reacting substance from the ground state to the transition state) .**
The enzyme is **not** modified during the reaction. The **initial** molecules are the **substrates** of the enzyme, and the molecules **formed** from these substrates are the **products** of the reaction.
Almost all metabolic processes in the cell require enzymes to run at a speed that is sufficient to sustain life.

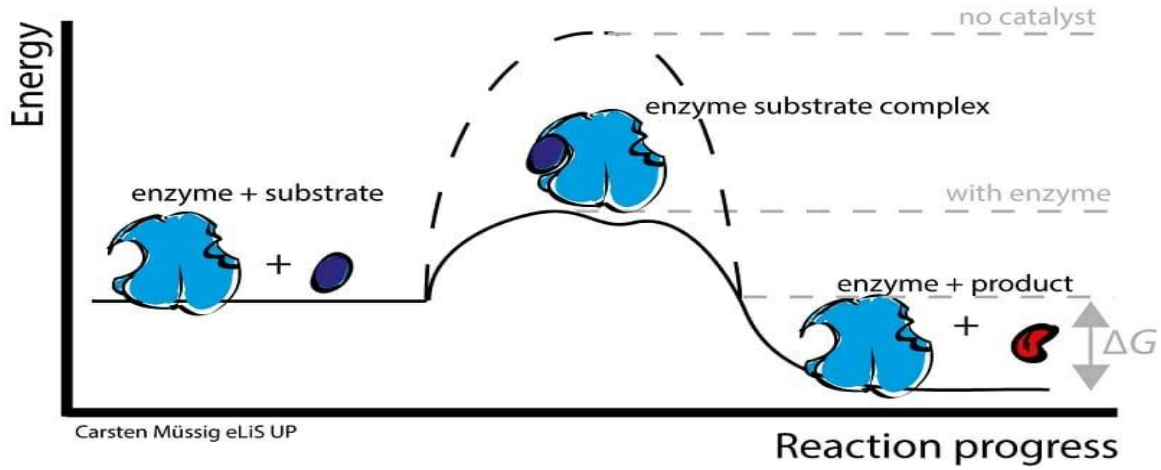
Enzymes are characterized by **two fundamental properties. First,** they increase the rate of chemical reactions without themselves being consumed or permanently altered by the reaction. **Second,** they increase reaction rates without altering the chemical equilibrium between reactants and products.
A molecule acted upon by an enzyme (as a substrate [S]) is converted to a product (P) as the result of the reaction. In the absence of the enzyme, the reaction can be written as follows:

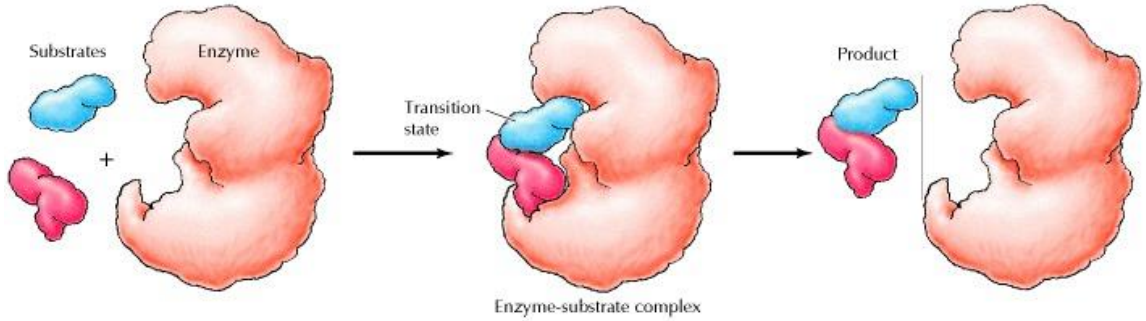


The chemical equilibrium between S and P is determined by the laws of thermodynamics and is represented by the ratio of the forward and reverse reaction rates (S→P and P→S, respectively).
In the presence of the appropriate enzyme, the **conversion of S to P is accelerated, but the equilibrium between S and P is unaltered.** Therefore, the enzyme must accelerate both the forward and reverse reactions equally. The reaction can be written as follows:



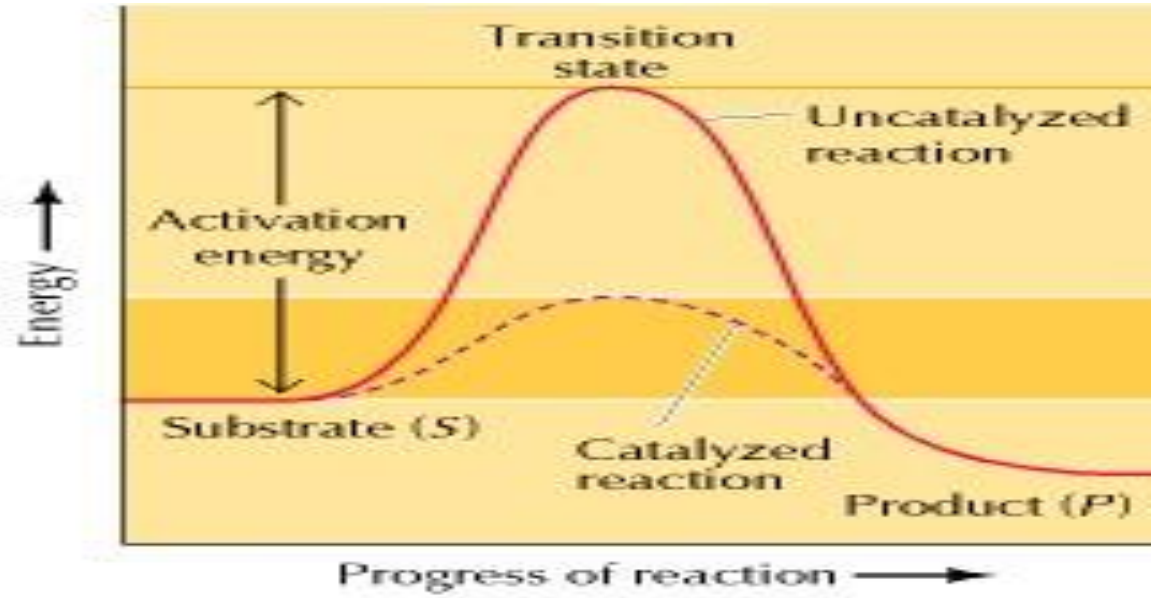
This is the role that enzymes play. They react with the substrate to form an **intermediate complex**—a "transition state"—that requires less energy for the reaction to proceed.
The unstable intermediate compound quickly breaks down to form reaction products, and the unchanged enzyme is free to react with other substrate molecules.





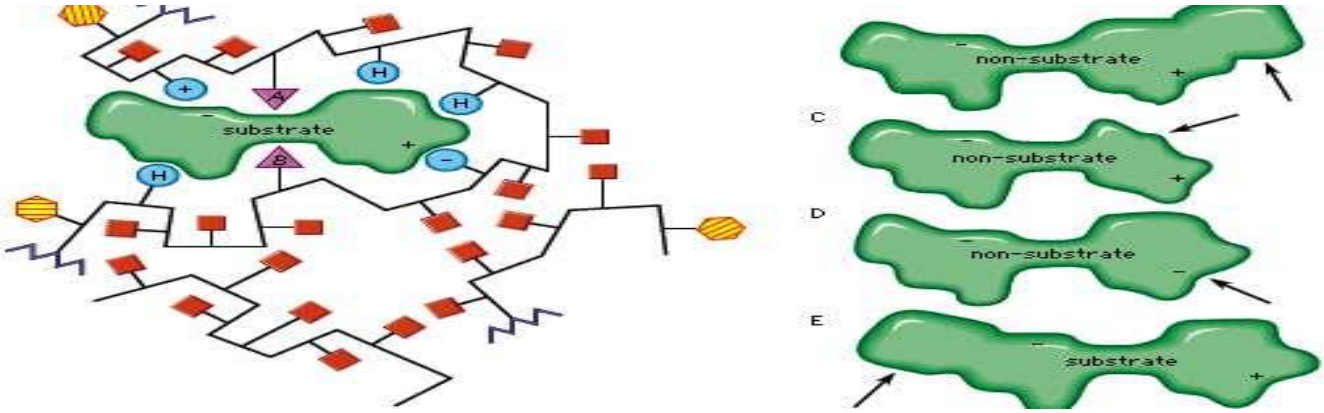
The effect of the enzyme on such a reaction is best illustrated by the energy changes that must occur during the conversion of *S* to *P*. The equilibrium of the reaction is determined by the final energy states of *S* and *P*, which are unaffected by enzymatic catalysis. In order for the reaction to proceed, however, the **substrate** must first be converted to a **higher energy state**, called the **transition state.** The energy required to reach the transition state (the activation energy) constitutes a barrier to the progress of the reaction, limiting the rate of the reaction.

Enzymes act by reducing the activation energy, thereby **increasing the rate of reaction.** The increased rate is the same in both the forward and reverse directions, since both must pass through the same transition state.



Only a certain region of the enzyme, called the active site, binds to the substrate, which is a groove or pocket formed by the folding pattern of the protein.

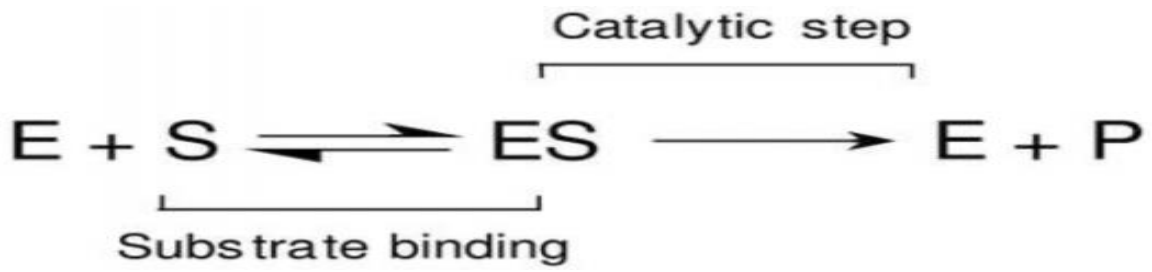
This three-dimensional structure, together with the chemical and electrical properties of the amino acids and cofactors within the active site, permits only a particular substrate to bind to the site, thus determining the enzyme's specificity.



The catalytic activity of enzyme involves the binding of their substrates to form an **enzyme- substrate complex (*ES*)**

The substrate binds to a **specific region(active site),** then the **substrate** is **converted into the product** of the reaction, which is then **released** from the enzyme

The active site is the region of an enzyme that binds substrates and catalyses an enzymatic reaction
The enzyme-catalysed reaction can thus be written as follows:



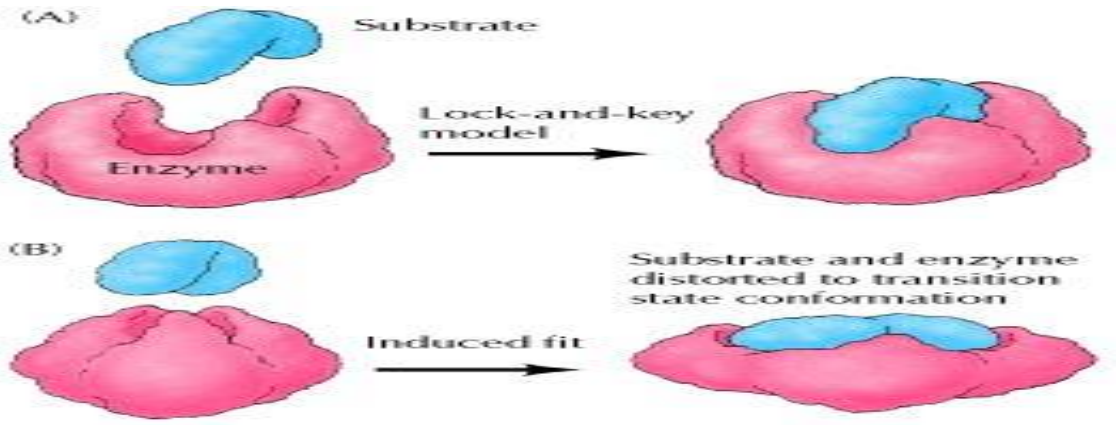
Note that **E appears unaltered** on both sides of the equation, **so the equilibrium is unaffected**.
However, the enzyme provides a surface upon which the reactions converting *S* to *P* can occur more readily.
This is a result of interactions between the enzyme and substrate that lower the energy of activation and favour formation of the transition state. Because the final energy state of *P* is lower than that of *S*, the reaction proceeds from left to right.

Mechanisms of Enzymatic Catalysis

The binding of a substrate to the active site of an enzyme is a very **specific interaction. The Active sites** are clefts or grooves on the surface of an enzyme, usually composed of **amino acids** from different parts of the polypeptide chain that are brought together in the tertiary structure of the folded protein(The three-dimensional folding of a polypeptide chain that gives the protein its functional form). Substrates bind to the active site by **noncovalent interactions,** including **hydrogen bonds, ionic bonds, and hydrophobic interactions**, when a substrate is bound to the active site of an enzyme, multiple mechanisms can accelerate its conversion to the product of the reaction.

Most biochemical reactions involve interactions between two or more different substrates. For example, the formation of a peptide bond involves the **joining of two amino acids.**
The enzyme provides a template upon which the reactants are brought together and properly oriented to favour the formation of the transition state in which they interact.

Enzymes accelerate reactions by altering the conformation of their substrates to approach the transition state. The simplest model of enzyme- substrate interaction is the **lock-and-key model**, in which the substrate fits precisely into the active site . The configurations of both the enzyme and substrate are modified by substrate binding a process called **induced fit**.
The conformation of the substrate is altered so that it more closely resembles that of the transition state, which is stabilized by its tight binding to the enzyme, thereby lowering the required energy of activation.



Models of enzyme-substrate interaction :

A) In the lock-and-key model, the substrate fits precisely into the active site of the enzyme as the key fits into the lock and hence it is called the lock and key model. Thus the active site by itself provides a rigid , pre-shaped template fitting with size and shape of the substrate molecule . This model proposes that substrate binds with rigid pre-existing template of the active site , provides additional groups for binding other ligands.

(B) In the induced-fit model, substrate binding distorts the conformations of both substrate and enzyme. This distortion brings the substrate closer

to the conformation of the transition state, thereby accelerating the reaction. The important feature of this model is flexibility of the region of active site. According to this , active site does not possess a rigid , performed structure on enzyme to fit the substrate .On the contrary , the substrate during its binding induces conformational changes in the active site to attain the final catalytic shape and form.

The part of the enzyme where the substrate binds is called **the active site**. Enzymes are highly specific and will only bind one type of substrate, this is because an enzyme and substrate both possess specific **complementary geometric shapes** that fit exactly into one another.
This is often referred to as "**the lock and key**" model. An enzyme only fits one particular kind of substrate in the same way that a lock only fits one particular kind of key.

