3

ENZYMES

After completing this lesson, you will be able to

- Describe the structure of enzyme.
- Explain the role and component parts of the active site of an enzyme.
- Differentiate among the three types of co-factors i.e. in organic ions, prosthetic group and co-enzymes, by giving examples.
- Explain the mechanism of enzyme action through Induced Fit Model, comparing it with Lock and Key Model.
- Explain how an enzyme catalyzes specific reactions.
- Define energy of activation and explain through graph how an enzyme speeds up a reaction by lowering the energy of activation.
- Describe the effect of temperature on the rate of enzyme action
- Compare the optimum temperatures of enzymes of human and thermophilic bacteria.
- Describe the range of pH at which human enzymes function
- Compare the optimum pH of different enzymes like trypsin, pepsin, pepane.
- Describe how the concentration of enzyme affects the rate of enzyme action.
- Explain the effect of substrate concentration on the rate of enzyme action.
- Construct and interpret graphs based on data about the effect of temperature, enzyme concentration and substrate concentration on the rate of enzyme action.
- Describe enzymatic inhibition, its types and its significance.
- Name the molecules which act as inhibitors.
- Categorize inhibitors into competitive and non-competitive inhibitors.
- Explain feedback inhibition.
- Classify enzymes on the basis of the reactions catalyzed (oxido-reductases, transferases, hydrolases, hydrolyases, isomerases, and ligases).
- Classify enzymes on the basis of the substrates they use (lipases, diastase, amylase, proteases etc).

You got a brief introduction about enzymes in IX-X biology course. There is complete check and balance on the chemistry of cell, which is exhibited through various enzymatic reactions going on within a cell. The concepts developed in this chapter will construct knowledge where you will be able to analyze comprehend and apply that knowledge.

The sum of all the chemical reactions going on in a cell is known as **metabolism**. These reactions have to be carried out very quickly so that their products can be utilized in various life activities in the cells. **Enzymes** are biological catalysts and therefore they speed up the biochemical reaction without being consumed.

Some common properties of enzymes are:

(i) Increase the speed of chemical reaction.

(ii) Required in very small quantity for the reaction.

(iii) Highly sensitive to pH and temperature.

(iv) Either highly specific or slightly less specific.

(v) Can work in *vivo* (living cells) as well as in *vitro* (glassware).

(vi) Some require co-factor for proper activity.

(vii) Lower the need of activation energy.

(viii) Only speed up a reaction and do not affect the equilibrium of the reaction.

Science Titbits

During the early nineteenth century, two French chemists, **Payen** and **Persoz** grounded up barley seeds in water to make a crude mixture that would digest starch. They gave the name **diastase** whatever it was that digested the starch.

3.1 ENZYME STRUCTURE

With exception of ribozymes, all the enzymes are globular proteins which are made up of one or more polypeptides. **Ribozymes** are the enzymes which consist of RNA and are found in ribosomes. For example, peptidyl transferase is a ribozyme which forms peptide bond during protein synthesis.

3.1.1 Shape of Enzymes and Components of an Active Site

Majority of enzymes which are protein in nature can have molecular weights ranging from about 10,000 to over 1 million. Such enzymes have tertiary or quaternary structures. The catalytic activity of an enzyme is located in its **active site** which is a specific charge bearing, three dimensional cavity. The substrate (the reactant which is to be converted into product) molecule is attached to the active site by non-covalent interactions like hydrogen bonding and hydrophobic interactions. Active site consists of 3-12 amino acids which may be scattered in the polypeptide but are brought together in a particular fashion due to secondary and tertiary folding of the protein molecule, e.g., the active site for aldolase consists of glycine, histidine, and alanine amino acids. An active site consists of two functional regions, i.e., binding site and catalytic site. Some amino acids have active site which makes bonds with substrate constitute the **binding site** while the other amino acids which cause conversion of substrate

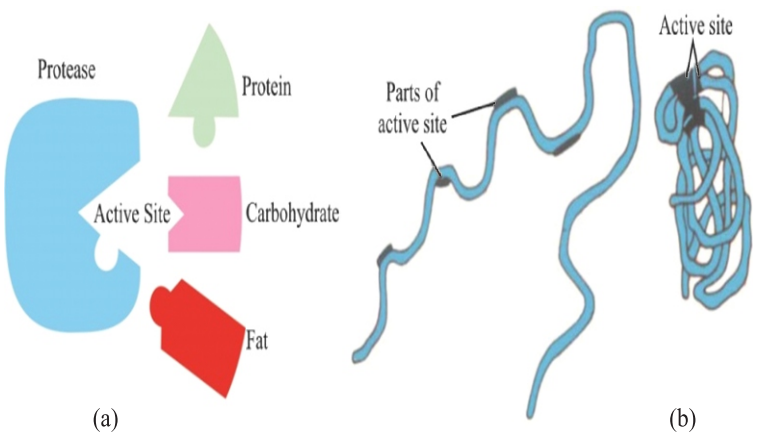


Fig: 3.1: Active site: (a) Which substrate fits the active site? (b) Grouping of amino acids of a polypeptide during the formation of tertiary structure to produce an active site.

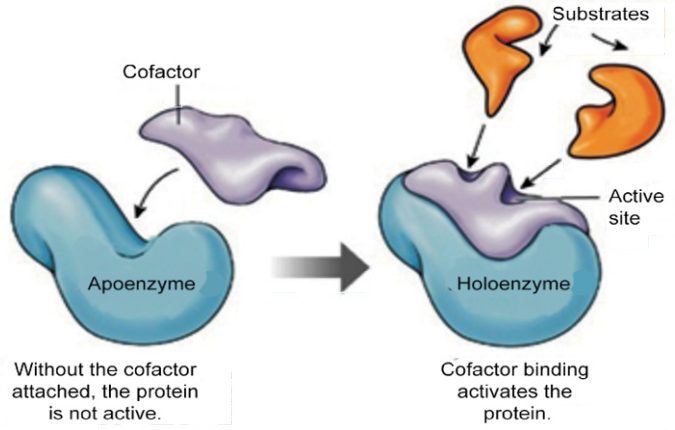


Fig. 3.2 Structure of enzyme

into product (catalysis) constitute the **catalytic site**. The shape of active site is designed according to the substrate therefore only a particular substrate can attach to the active site, however, sometime related substrate can also bind to the active site.

Some enzymes also require a non-protein part, the **cofactor** which is not only responsible for the attachment of substrate to the active site but also participate in catalytic process. The final shape of active site is actually established after the attachment of cofactor. An enzyme which requires a cofactor becomes active only if the cofactor is combined with it. Such an active enzyme is called **holoenzyme**. If the cofactor is not available the remaining protein part of enzyme becomes catalytically inactive and is called **apoenzyme**. On the other hand, the enzymes which do not require cofactor can also show active and inactive states. Pepsin is an example of such enzyme. It is secreted by gastric gland from stomach wall in an inactive state, the **pepsinogen**. In this state, it has an additional polypeptide fragment attached to its active site which does not allow the binding of substrate, hence it remains inactive. When pepsinogen is exposed in HCl (as in stomach cavity) the additional polypeptide fragment is removed and as a result inactive (apoenzyme) pepsinogen is changed into its active (holoenzyme) form, the **pepsin.**

Science Titbits

How are enzymes formed? Enzymes are proteins, so they are formed as per message or base sequence in DNA. Enzymes are synthesized by living cells but they retain their catalytic action even when extracted from cells, i.e., they can act in vitro. These days' enzymes are also being produced by recombinant DNA technology.

3.1.2 Types of Cofactors

The cofactor may be inorganic or organic molecules. The inorganic cofactors are different metallic ions such as Fe^{++}, Mg^{++}, Cu^{++}, Zn^{++}, etc. These are only attached to the enzymes when substrate gets bind i.e., they are detachable cofactors. Such cofactors are also called **activators**.

Glucose + ATP $\xrightarrow{\text{Hexokinase + } Mg^{+2}}$ **Glucose-6-phosphate + ADP**

The organic cofactors are either co-enzymes or prosthetic groups. The **coenzymes** are the derivatives of vitamins. For example ATP, NAD^+, FAD^+ are common coenzymes. Like inorganic cofactors they are also attached to the enzymes when substrate gets bind i.e., they are also detachable cofactors.

Ethyl alcohol $\xrightarrow{\text{Alcohol dehydrogenase + } NAD^+}$ **Acetaldehyde + $NADH_2$**

On the other hand a **prosthetic group** is covalently bonded part of an enzyme which is permanently attached to enzyme and does not detach after the completion of reaction. An iron containing porphyrin ring attached to some enzymes like cytochromes is the example of prosthetic group.

3.2 MECHANISM OF ENZYME ACTION

In an enzyme-catalysed reaction, the substrate first binds to the active site of the enzyme to form an **enzyme-substrate (ES) complex**, then the substrate is converted into **product** while it is attached to the enzyme **(EP complex)**, and finally the product is released, thus allowing the enzyme to start all over again.

$$\underset{\text{enzyme}}{E} + \underset{\text{substrate}}{S} \longrightarrow \underset{\text{enzyme-substrate complex}}{ES} \longrightarrow \underset{\text{enzyme-product complex}}{EP} \longrightarrow \underset{\text{enzyme}}{E} + \underset{\text{product}}{P}$$

Actually, the enzyme can make the local conditions inside the active site quite different from those outside (such as pH, water concentration, charge), so that the reaction is more likely to happen. For example, if a substrate is to be split, a bond might be stretched by the enzyme, making it more likely to break.

3.2.1 Models of Enzyme Action

The mechanism of enzyme action can be explained with the help of two different models. **Emil Fischer** proposed **Lock and key model** (in 1894). According to this model the active site of the enzyme has definite shape and rigid structure. Shape of active site is complementary to the shape of substrate. Therefore, a particular substrate can only bind to the active site. The active site remains unchanged during or after the reaction. Lock and key model assumes that like a particular key opens a particular lock, a specific **enzyme (key)** acts upon a particular **substrate (lock).** Actually, the notched portion of the key is equivalent to the active site on the enzyme. It reflects that enzymes are highly specific in their action and each enzyme can carry out only one particular reaction. The enzymes,

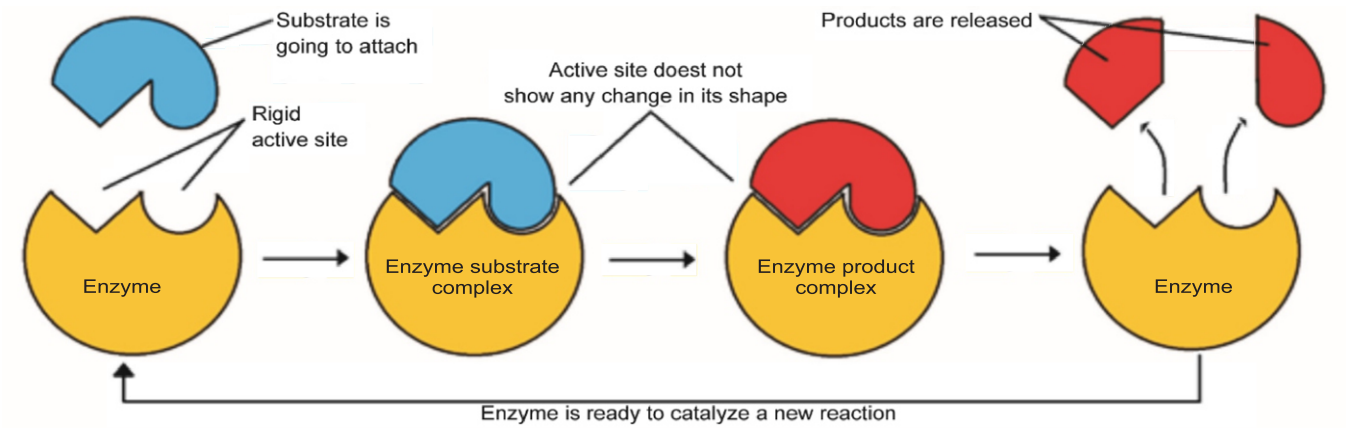


Fig: 3.3: Fischer's "Lock and Key" hypothesis of enzyme action

which work according to this model, are called **non-regulatory enzymes**. However, this model is exercised by a very small number of enzymes, for example sucrase, maltase etc. The ability of enzyme to catalyze one specific reaction is perhaps its most significant

property. Although, many enzymes show a broad range of specificity towards the substrate they catalyze. When one enzyme can catalyze only one substrate and essentially no others it is called **absolute specificity** e.g., urease.

Urea + Water $\xrightarrow{\text{Urease}}$ **Ammonia + Carbon dioxide**

Koshland proposed **Induced fit model** (in 1959). According to this model the active site is flexible; therefore, it is modified as the substrate interacts with enzyme. The amino acids, which makeup the active site are molded into a precise shape which enables the enzyme to perform its catalytic function more effectively. The change which is induced in the shape of active site is responsible for the conversion of substrate into product. As the reaction is completed the active site regains its original shape. This is the flexibility of active site which allows more than one type of related substrates to be attached on active site and therefore, an enzyme can carry out more than one type of related reactions. The example is carbonic anhydrase which can add O_2 to haemoglobin as well as can control the formation of carbonic acid and bicarbonates in blood.

Enzymes, which follow the induced fit mechanism, are called **regulatory** or **allosteric enzymes** for example hexokinase.

$$Hb + 4O_2 \xrightarrow{\text{Carbonic anhydrase}} Hb4O_2 \text{ (Oxyhaemoglobin)}$$

$$CO_2 + H_2O \xrightarrow{\text{Carbonic anhydrase}} H_2CO_3 \xrightarrow{\text{Carbonic anhydrase}} H^+ + HCO^-$$

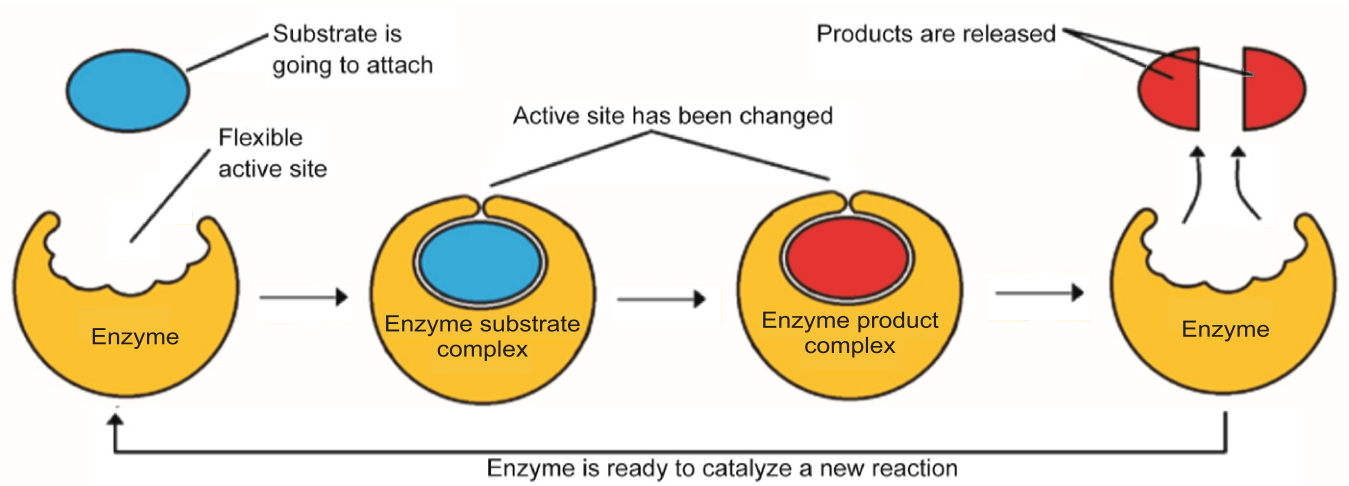


Fig: 3.4: Koshland's "Induced Fit" model of enzyme action

3.2.2 Energy of Activation

Molecules do not react with one another unless they are activated in some way. The energy that must be added to cause molecules to react with one another is called the **energy of activation**. In nonliving system we use heat as energy of activation to increase the number of effective collision between molecules. In living systems large amount of heat cannot be used as energy of activation. Why? All living cells and organisms are

mainly composed of temperature sensitive protein molecules. About 1,000 chemical reactions are being carried out in a cell at any time. Energy of activation required for such a large number of reactions cannot be provided by living system.

The living system works in isothermal condition. The excited state of molecules or reactants is achieved by biochemical process. Enzyme (E) reacts with reactant (A) to form an AE transitional complex. The energy level of AE complex reaches to the energy level of reactant B. AE complex then reacts with reactant B to form AB and enzyme (E) is released.

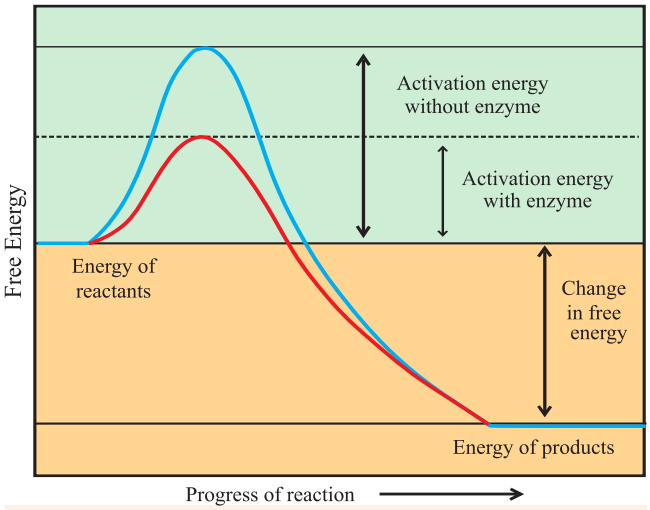


Fig: 3.5: Energy of activation: Enzymes speed the rate of chemical reactions because they lower the amount of energy required to activate the reactants and lower the need of activation energy

A + E → AE Complex + B → AB + E

Enzyme does decrease the energy of activation by changing energy dependent process to energy independent process. Thus the energy of activation is "energy required to break the existing bonds and begin the reaction". An enzyme greatly reduces the activation energy necessary to initiate a chemical reaction.

3.3 FACTORS AFFECTING THE RATE OF ENZYMATIC ACTION

The rate of enzymatic reaction is measured by the amount of substrate changed or amount of product formed, during a period of time. The external conditions which affect rate of enzyme reactions are: temperature, pH, concentration of enzyme and substrate concentration.

3.3.1 Temperature

Heating increases molecular motion. Thus the molecules of the substrate and enzyme move more quickly, so probability of a reaction to occur is increased. Increasing temperature affect the rate of reaction in such a way that an increase of just 10°C in the existing temperature doubles the rate of reaction but this effect remains up to a certain limit. The temperature that promotes maximum activity is called an **optimum temperature**. If the temperature is increased above this level, then a decrease in the rate of the reaction occurs despite the increasing frequencies of collision. This is because the secondary and tertiary structures of the enzyme have been disrupted

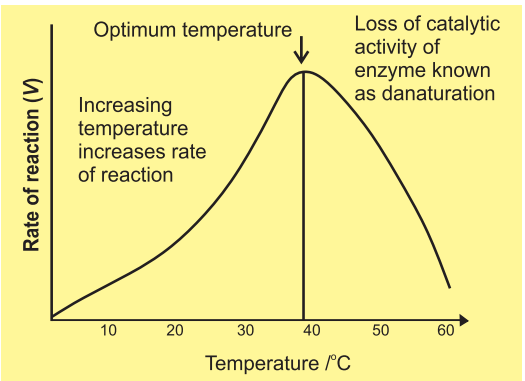


Fig: 3.6 (a): Effect of temperature on the rate of an enzyme controlled reaction

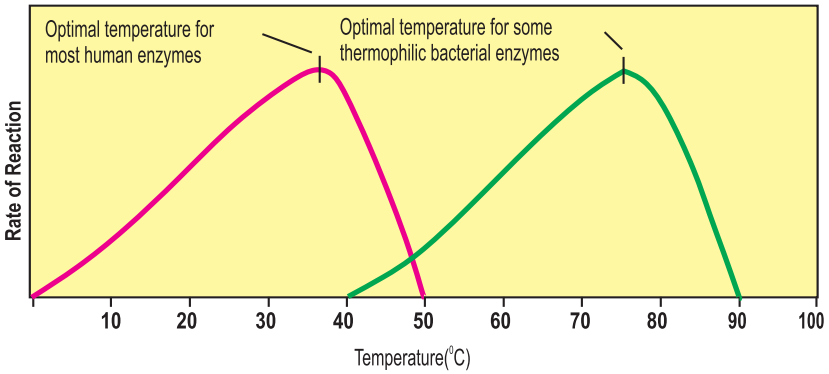


Fig: 3.6 (b): Optimum temperature for human enzymes and thermophilic bacteria

and the enzyme is said to be denatured. The enzyme unfolds and the precise structure of the active site is gradually lost. This temperature which causes denaturation of enzyme is called **maximum temperature**. The bonds which are most sensitive to temperature change are hydrogen bonds. All human enzymes have a optimum temperature of about 37-38°C, but bacteria living in hot springs may have an optimum temperature of 70°C or higher. Such enzymes have been used in biological washing powders for high temperature washes. If temperature is reduced to near or below freezing point, enzymes are inactivated, not denatured. They will regain their catalytic influence when higher temperatures are restored. This temperature where an inactive enzyme becomes active again is called **minimum temperature.**

3.3.2 pH

Every enzyme functions most effectively over a particular pH range. This narrow range of pH at which the maximum rate of reaction is achieved is called **optimum pH.** Enzyme conformation is sensitive to pH changes because pH influences the charges on the amino acid side chains that are involved in maintaining tertiary and quaternary structure of enzyme. Slight change in optimum pH of an enzyme causes ionization of amino acid of the enzyme therefore, they become inactive temporarily. On the other hand, extreme changes in optimum pH alter the ionic charge of the acidic and basic groups of enzyme and therefore disrupts the ionic bonding (denaturation) that helps to maintain the specific shape of the enzyme.

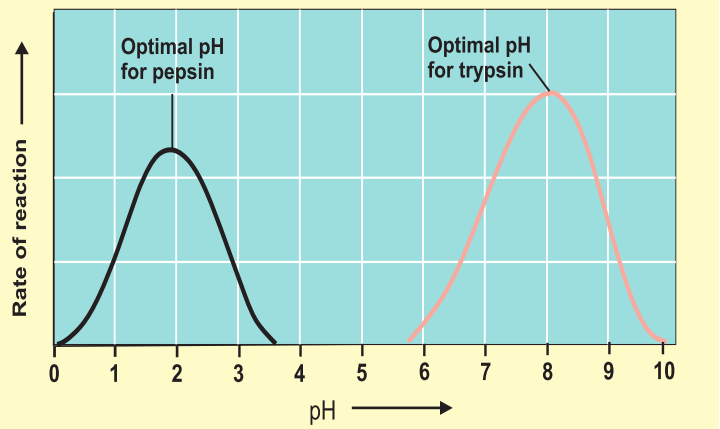


Fig: 3.7: Effect of pH on the rate of enzyme-controlled reaction

The optimum pH values for most enzymes fall in the range of pH 6-8, but there are exceptions. Some enzymes like papain from green papaya act both in acidic and alkaline media. Protein digesting enzyme pepsin is active in acidic medium at pH 2 and trypsin is inactive at this pH but shows maximum activity in alkaline medium at pH 8.

Critical Thinking

Industrial pollution can change the pH of a pond, lake or river to make the water more acidic. How can this affect the metabolic pathways of the plants that live in water?

3.3.3 Enzyme Concentration

Provided that the substrate concentration is maintained at a high level (unlimited availability), and other conditions such as pH and temperature are kept constant, the rate of reaction becomes directly proportional to the enzyme concentration. If there is only one enzyme in the system it can convert hundreds of substrates into products but it takes more time. By increasing concentration of enzyme, numbers of active sites become more available and the rate of conversion of substrate into product becomes fast. Such effect persists till the equilibrium state (when concentration of enzyme and substrate becomes equal), after that further increase in enzyme concentration will have no effect upon rate of reaction.

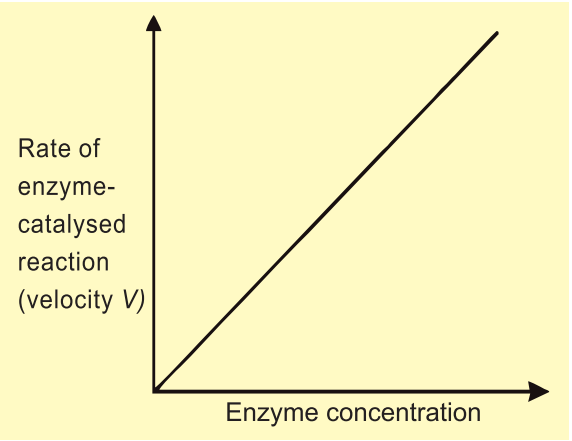


Fig: 3.8: Relationship between Enzyme concentration and the rate of an Enzyme-controlled reaction

3.3.4 Substrate Concentration

When other conditions such as pH and temperature are kept constant and the enzyme concentration is maintained at a higher level (unlimited availability), the increase in substrate concentration (S) increases the velocity (V) of the enzymatic reaction at first. The reaction ultimately reaches a maximum velocity at equilibrium state. The rise in V is decreased progressively with further increase in S. The reaction does not increase by any further rise in substrate concentration. This happens because all the active sites of enzyme molecules are occupied by the substrates (saturation) and no enzyme is left free to bind with additional molecules of the substrate.

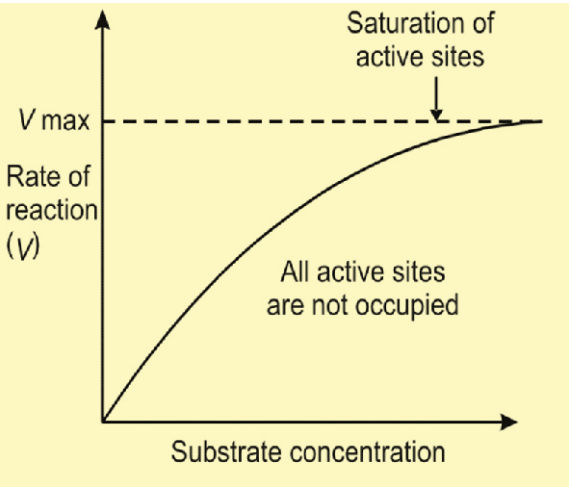


Fig: 3.9: Effect of Substrate concentration on the rate of Enzyme-controlled reaction

3.4 ENZYME INHIBITION

The phenomenon in which an enzyme fails to catalyze a reaction is called **enzyme inhibition** and the molecules which react with enzyme but are not converted into desired products are called **enzyme inhibitors**. In general, the enzyme inhibition is a normal part of the regulation of enzyme activity within cells but sometimes when external factors cause enzyme inhibition; it may become dangerous for life. The molecules which act as inhibitors include poisons, cyanides, antibodies, anti-metabolites, penicillin, sulpha drugs etc. Inhibition may be competitive or noncompetitive.

Science Titbits

Penicillin blocks the active site of an enzyme unique to bacteria. When penicillin is taken, bacteria die but human are unaffected.

3.4.1 Competitive Inhibition

A type of enzyme inhibition in which enzyme activity is blocked by the presence of a chemical that compete with the substrate for binding to the active site is called **competitive inhibition**. Usually a competitive inhibitor is structurally similar to the normal substrate and so fits into the active site of the enzyme. However, it is not similar enough to substitute fully for the normal substrate in the chemical reaction and the enzyme cannot catalyze it to form reaction products. Competitive inhibition is usually temporary, and the inhibitor eventually leaves the enzyme hence it is also called **reversible inhibition**. This means that the level of inhibition depends on the relative concentrations of substrate and i nhibitor, since they are competing for places in enzyme active sites. Therefore, if the concentration of the substrate is increased relative to the concentration of the inhibitor, the active site will usually be occupied by the substrate. An example of inhibitor is **malonate**. Succinate dehydrogenase that catalyzes the formation of fumarate from succinate is competitively inhibited by malonate.

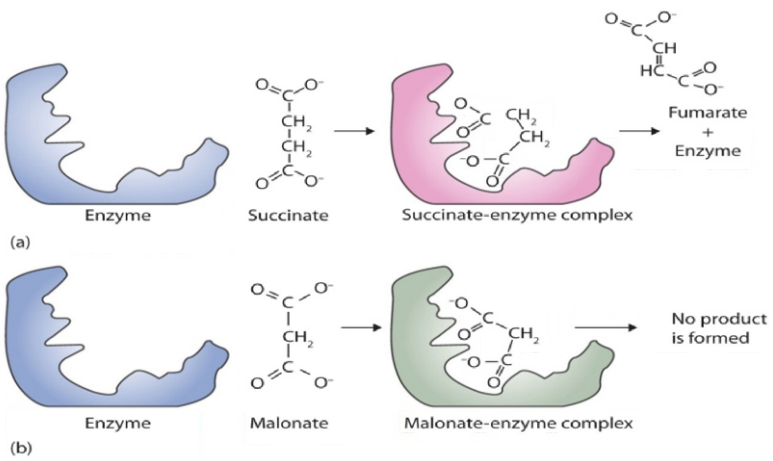


Fig: 3.10: Effect of malonate as competitive inhibitors

The **importance** of competitive inhibitors is: (a) It supports lock and key hypothesis. (b) It shows that substances which are similar to substrate are not acted upon by enzymes. (c) Competitive inhibitors are used as drugs in the control of bacterial pathogens. Antibiotics known as sulphonamides are used to combat bacterial infection.

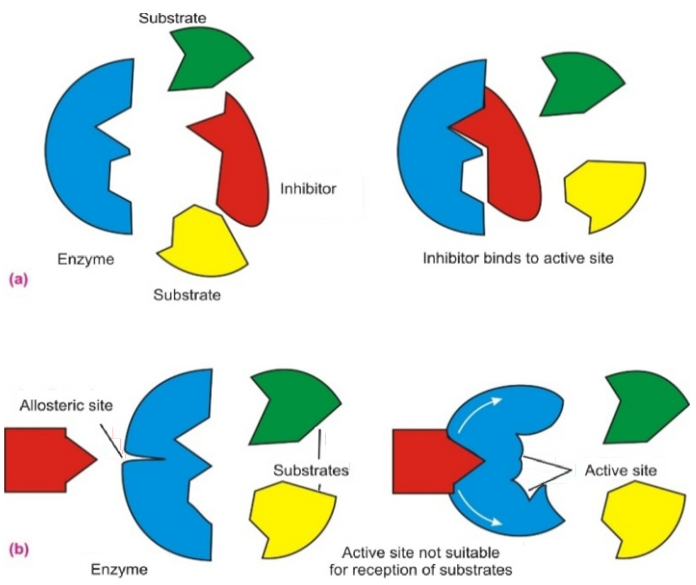


Fig: 3.11: (a) Competitive inhibition (b) Non-competitive inhibition

3.4.2 Non-Competitive Inhibitors

In non-competitive inhibition the inhibitor molecule binds to an enzyme other than active site. The other binding site of enzyme is called **allosteric site.** The non-competitive inhibitors inactivate the enzyme temporarily (reversible inhibition) or they denature the enzyme permanently (irreversible inhibition). **Reversible non-competitive** enzyme inhibitors work not by preventing the

formation of enzyme-substrate complexes, but by preventing the formation of enzyme-product complexes. So they prevent the substrate to be converted into product. Feedback inhibition is an example of reversible non-competitive enzyme inhibition

On the other hand, an **irreversible non-competitive** enzyme inhibitor destroys enzyme by altering its shape so that the substrate cannot bind to the active site. The examples of irreversible non-competitive inhibitors include cyanides and salts of heavy metals. **Cyanides** are potent poisons of living organism because they can kill an organism by inhibiting cytochrome oxidase essential for cellular respiration. They block the action of these enzymes by combining with iron which may be present in the prosthetic group. **Ions of heavy metals** such as mercury, silver and copper (Hg^{++}, Ag^{+}, and Cu^{++}) combine with thiol (-SH) groups in the enzyme breaking the disulphide bridges. These bridges are important in maintaining tertiary structure. When these bridges are broken, the enzyme becomes denatured and inactive.

Critical Thinking

Suggest why substrate concentration has no effect on non-competitive inhibition?

3.4.3 Feedback Inhibition

The activity of almost every enzyme in a cell can be regulated by its product. When the activity of an enzyme is inhibited by its own product, it is called feedback inhibition. This is a type of reversible non-competitive inhibition. This phenomenon is a part of normal regulatory mechanism and usually happens during the regulation of metabolic pathways. For example, the amino acid aspartate becomes the amino acid threonine by a sequence of five enzymatic reactions. When threonine, the end product of this pathway, is present in excess, it binds to an allosteric site on enzyme 1 on this pathway and then the active site is no longer able to bind aspartate. When all the threonine is consumed in cellular events, the threonine molecule which is attached to the allosteric site is also removed; the pathway resumes its activity once again.

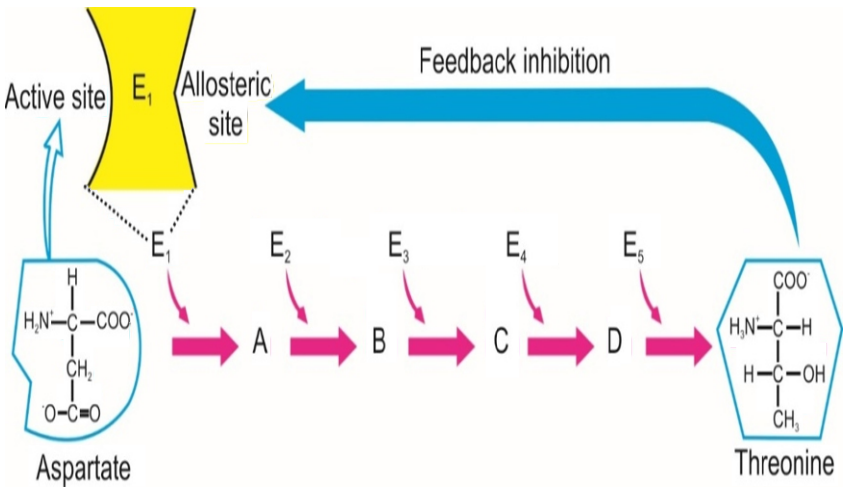


Fig: 3.12: Feedback inhibition

Skills: Analyzing

- **Identify the competitive and non-competitive inhibitors from the given list of chemical (consult any book of Biochemistry or Enzymology).** (Answer is given below)

Competitive inhibitors: Antibodies, antimetabolites, penicillin, iodoacetate, melonate, CoA (high concentration).

Non-competitive inhibitors: Acetaldehyde Di-Isopropyl fluorophosphate (DFP- nerve gas), mercury, silver, copper, cyanide.

3.5 CLASSIFICATION OF ENZYMES

Enzymes can be classified either on the basis of reaction types that they catalyze or on the basis of substrate which are acted upon by the enzyme.

3.5.1 Classification based upon reaction type

A systematic nomenclature and classification of enzymes based on reaction types and reaction mechanism was given by International Union of Biochemistry (in 1961).

On that basis all the enzymes have been classified into six groups:

1. Oxidoreductases
2. Transferases
3. Hydrolases
4. Lyases
5. Isomerases
6. Ligases

1- Oxidoreductases

These enzymes catalyze oxidation/reduction of their substrate and act by removing or adding electron or H^+ ions from or to the substrate. For example **cytochrome oxidase** oxidizes cytochrome.

2- Transferases

These enzymes catalyze the transfer of specific functional group other than hydrogen from one substrate to another. The chemical group transferred in the process is not in a free state, for example **hexokinase** transfers a phosphate group from ATP to glucose.

Science Titbits

How are enzymes named?

(a) Enzymes are named by adding "ase" to the name of substrate they act, e.g., proteases, lipases etc. (b) Enzymes are named according to the types of reaction they catalyse, e.g., oxidases, reductases etc. (c) Enzymes are named by taking into consideration both the substrate acted upon and the type of reaction catalysed, e.g., DNA- polymerase. (d) Some enzymes are named as per substance synthesized, e.g., rhodonase catalyses synthesis of rhodonate from hydrochloric acid and sodium thiosulphate.

3- Hydrolases

These enzymes bring about the breakdown of large complex organic molecules into smaller ones by adding water (hydrolysis) and breaking the specific covalent bonds. Examples are proteolytic enzymes which breakdown proteins into peptones and peptides such as **pepsin, renin** and **trypsin**. Other digestive enzymes that work in digestive tract are also the examples of hydrolases.

4- Lyases

These enzymes catalyze the breakdown of specific covalent bonds and removal of groups without hydrolysis. For example **histidine decarboxylase** breaks the covalent bonds between carbon atoms in histidine forming carbon dioxide and histamine.

5- Isomerases

These enzymes bring about intra-molecular rearrangement of atoms in the molecules and thus forming one isomer from another. For example **phosphohexose isomerase** changes glucose 6- phosphate to fructose 6- phosphate.

6- Ligases (Synthetases)

These enzymes bring about joining together of two molecules. The energy is derived by hydrolysis of ATP. For example **polymerases** are responsible for linking monomers into a polymer such as DNA or RNA.

Table 3.1:Classifiation of enzymes based upon reaction type

Sr. No	Enzyme Class	General Scheme of Reaction
1.	Oxidoreductases	$A_{red} + B_{ox} \rightleftharpoons A_{ox} + B_{red}$
2.	Transferases	$A—B + C \longrightarrow A + C—B$
3.	Hydrolases	$A—B + H_2O \longrightarrow A—H + B—OH$
4.	Lyases	$A—B \rightleftharpoons A + B$ (reverse reaction syntheses)
5.	Isomerases	$A—B—C \rightleftharpoons A—C—B$
6.	Ligases (synthetases)	$A + B + ATP \longrightarrow A—B + ADP + Pi$

3.5.2 Classification based upon substrate

Enzymes can be classified on the basis of substrates they use. Some of the examples are: proteases, lipases, carbohydrases and nucleases.

1- Proteases

These enzymes act upon proteins. Examples are: **pepsin** and **trypsin** (both digest large polypeptides into small polypeptides or peptones), **aminopeptidases** and **carboxypeptidases** (both digest peptones into dipeptides) and **erypsin** (digest dipeptides into amino acids)

2- Lipases

These enzymes hydrolyze lipids into fatty acids and glycerols. Examples are **pancreatic lipases.**

3- Carbohydrases

These enzymes cause breakdown of carbohydrates. Examples are:

(a) **amylase** (digest starch or glycogen into maltose)

(b) **cellulase** (digest cellulose into cellubiose, a disaccharide)

(c) **maltase** (digest maltose into glucoses)

(d) **sucrase** (digest sucrose into glucose and fructose)

(e) **lactase** (digest lactose into galactose and glucose)

4- Nucleases

These are involved in the breakdown of DNA and RNA. Examples are:

(a) **RNAases** (digest RNA into ribonucleotides)

(b) **DNAases** (digest DNA into deoxyribo nucleotides).

(c) **ATPases** (cause hydrolysis of ATP in muscles etc.)

Science, Technology and Society Connections

- **List the diagnostic uses of enzymes.**

(a) Aldolase: progressive muscular dystrophy, viral hepatitis and advanced cancer of the prostate
(b) Creatine Phosphokinase: damage to muscle cells.
(c) Gamma-glutamyl Transpeptidase: in assessing liver function.
(d) Lactic Dehydrogenase: in differentiating heart attack, anemia, lung injury, or liver disease.
(e) Lipase: Damage to the pancreas.

Science, Technology and Society Connections

- **Venoms as enzyme inhibitors**

Snake venom is highly modified saliva that is produced by special glands of certain species of snakes. Snake venom is a combination of many toxins (proteins) and different enzymes, use for the purposes like increasing the prey's uptake of toxins. Snake venom inhibits cholinesterase to make the prey lose control of its muscles. Venom is an inhibitor for an essential enzyme cytochrome oxidase in the cells. There are three distinct type of venom that act on the body differently.

(1) Hemotoxic venoms act on the heart and cardiovascular system.

(2) Neurotoxic venom acts on the nervous system and brain.

(3) Cytotoxic venom has a localized action at the site of the bite. Venom occupies the active site of the enzyme or combining with the iron which may present in the prosthetic group or which may be required as an enzyme activator.

Activity

1. Performing of chemical test to demonstrate that enzymes are proteins
2. Performing amylase test on starch with boiled amylase and un-boiled amylase in separate test tubes and confirmation through iodine test

Exercise

MCQs

1. Select the correct answer

(i) The catalytic activity of an enzyme is restricted to its small portion called

(A) active site (B) passive site

(C) regulation site (D) allosteric site

(ii) Which of the following has a coenzyme activity?

(A) NAD^+ (B) Ca^{++}

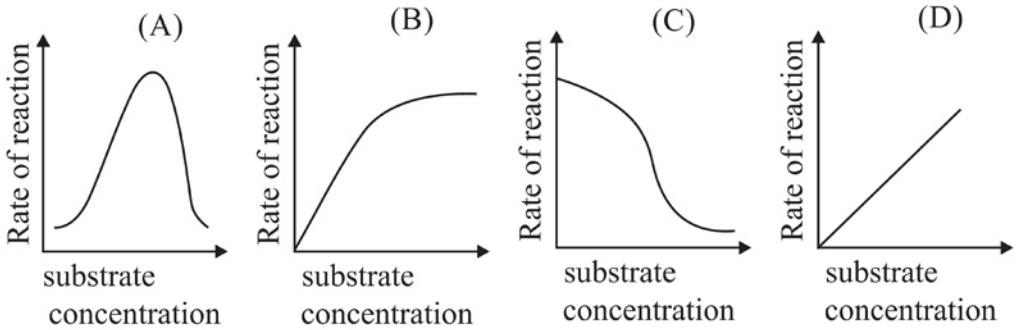
(C) both "a" and "b" (D) none of them

(iii) Non-competitive inhibitors react with enzymes at:

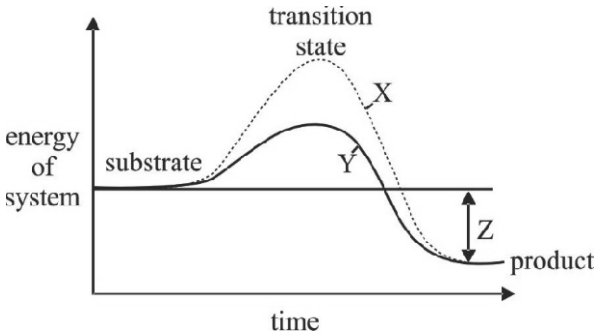
(A) active site (B) allosteric site

(C) both "a" and "b" (D) none of them

(iv) Which graph shows the expected relationship between enzyme activity and substrate concentration?



(v) The graph shows the effect of an enzyme on a reaction.



Which combination identifies X, Y and Z?

	X	Y	Z
A	catalyzed reaction	uncatalyzed reaction	activation energy
B	catalyzed reaction	uncatalyzed reaction	energy lost during reaction
C	uncatalyzed reaction	catalyzed reaction	energy gained by product
D	uncatalyzed reaction	catalyzed reaction	overall energy change

(vi) Combination of apoenzyme and coenzyme produces

(A) prosthetic group (B) holoenzyme

(C) enzyme (D) isoenzyme

(vii) The specificity of enzyme is due to their

(A) surface configuration (B) pH

(C) hydrogen bonding (D) high molecular weight

(viii) An essential feature of a competitive inhibitor is its ability to

(A) activate an operator gene (B) combine with prosthetic group

(C) modify a substrate (D) occupy an active site

(ix) The reaction rate of salivary amylase with starch decreases as the concentration of chloride ions is reduced. Which of the following describe the role of the chloride ions?

(A) allosteric inhibitors (B) cofactors

(C) coenzyme (D) competitive inhibitor

(x) How does an enzyme increase the rate of a reaction?

(A) by bringing the reacting molecules into precise orientation

(B) by increasing the rate of random collisions of molecules

(C) by shifting the point of equilibrium of the reaction

(D) by supplying the energy required to start the reaction

(xi) Many enzymes are secreted in inactive form to protect

(A) cell proteins (B) mitochondria

(C) cell membrane (D) cell DNA

(xii) Erypsin is an example of?

(A) carbohydrases (B) proteases

(C) lipases (D) nucleases

(xiii) Ribozymes consist of:

(A) only protein (B) protein + none protein part

(C) only RNA (D) none of them

Short Questions

2. What are ribozymes?
3. What is the structure of enzyme?
4. Explain the enzyme pepsin which does not require cofactor.
5. What is prosthetic group? Give an example.
6. What is the mechanism of enzyme action?
7. What is the role of free energy of activation in a chemical reaction?
8. List the external conditions which affect rate of enzyme reaction.
9. Compare the optimum temperatures of enzymes of human and thermophilic bacteria.
10. Describe the range of pH at which human enzymes function.

11. What are enzyme inhibitors? Name the molecules which act as enzyme inhibitors.
12. What is the importance of competitive enzyme inhibitors?
13. Describe cyanides as irreversible non-competitive inhibitor.
14. Describe ions of heavy metals as irreversible non-competitive inhibitor.
15. Write the difference between:
    (a) binding site and catalytic site of an enzyme
    (b) apoenzyme and holoenzyme
    (c) prosthetic group and coenzyme
    (d) inorganic cofactor and organic cofactor
    (e) lock and key model and Induced fit model of enzyme action
    (f) competitive and noncompetitive enzyme inhibitors
    (g) reversible non-competitive enzyme inhibitors and irreversible non-competivie enzyme inhibitors

Extensive Questions

16. Write the properties of enzymes.
17. Explain the role and component parts of the active site of an enzyme.
18. What are cofactors? Describe the two types of cofactors by giving examples.
19. Explain the mechanism of enzyme action through induced fit model.
20. Explain the mechanism of enzyme action through lock and key model.
21. Explain how an enzyme catalyzes specific reactions.
22. Explain through graph how an enzyme speeds up reaction by lowering the energy of activation.
23. Describe the effect of temperature on the rate of enzyme action.
24. Describe how the concentration of enzyme affects the rate of enzyme action.
25. Explain the effect of substrate concentration on the rate of enzyme action.
26. Describe enzymatic inhibition, its types and its significance.
27. Explain feedback mechanism with reference to enzymes.
28. Classify enzymes on the basis of reactions catalyzed.
29. Classify enzymes on the basis of the substrate they use.