Chapter 3

ENZYMES

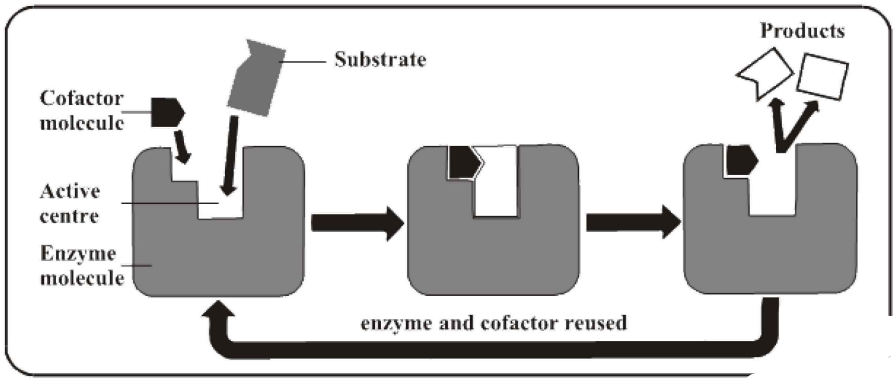
Q.1 Define enzyme, and write an account on enzymes.

Ans. **ENZYME**

"A protein which ***speeds up biochemical reaction*** without its used up is called enzyme".

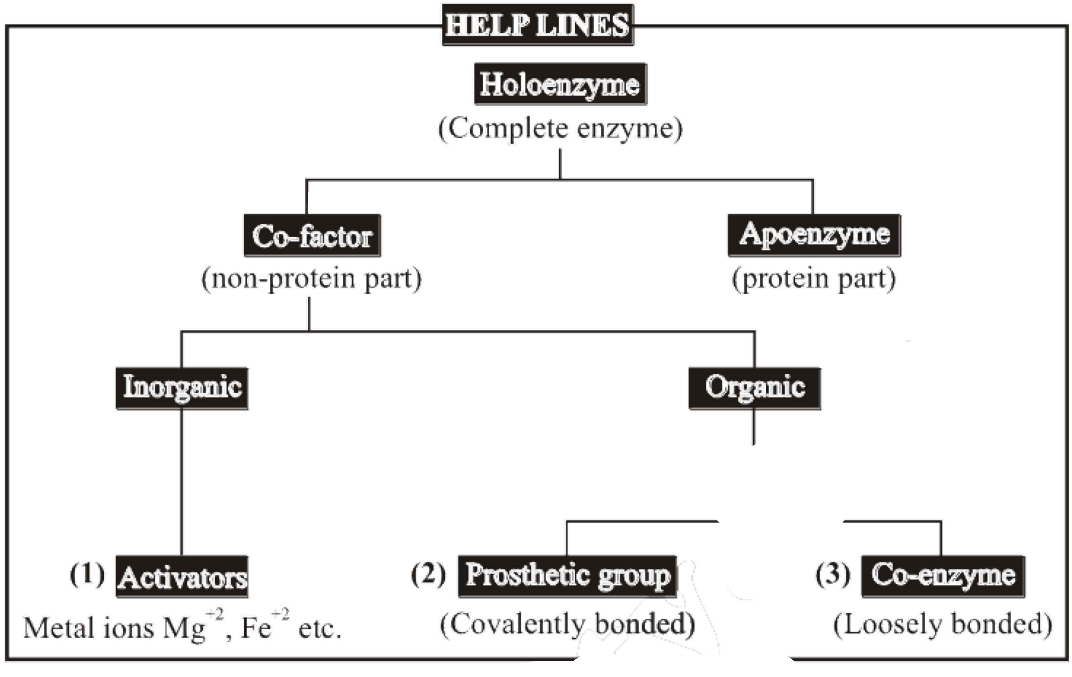
- Enzymes have very important role in life. It is that substance which is *not used up* in biochemical reaction.

  It is essential for proper activities of life, because without enzymes the reactions proceed very slow. *Proper speed* of reactions is essential for proper functions.
- The study of enzymes is called ***enzymology***
- General process of life like digestion, respiration, excretion etc. depend upon enzymes.
- Particular enzyme acts on a specific reactant or substrate. ***"Substrate is that substance on which enzyme acts".***
- There is a ***lock and key relationship*** between substrate and enzyme.
- Enzymes never become the part of product.
- Commonly the "suffix" of the enzyme is ***"__ase"***, in few case ***"__IN"*** is also ending of enzyme. *e.g., sucrase, maltase, lipase, catalase, amylase, orginase, enterokinase etc. and pepsin, trypsin, erepsin etc.*
- All enzymes are ***globular proteins.***



- **Co-Factors** and ***Coenzymes*** are necessary for catalytic activities of enzymes. Mg++, Fe++, Cu++ and Zn++ etc act as cofactors, but these are inorganic ions and termed as activators.
- **Coenzymes** organic molecules i.e. vitamins (NAD, Biotin) which also essential for reactions. The tightly bounded coenzyme situation is prosthetic group.
- ***Inactive Protein Portion of Enzyme:*** The remaining protein portion after removal of cofactor or prosthetic group is known as *apoenzyme*.

  Active Complex: Enzyme plus cofactor become the active complex, which is termed as holoenzymes.
- **Active Sites:** The site at which the substrate of an enzyme is bound during catalysis. Once bound, the substrate reacts to form a product or products, which are then released from the active site.
- ***Specific Enzymes and Specific Sites:*** Specific enzymes are produced on specific site, according to requirement. The enzymes for cellular respiration are found in *mitochondria*. *Ribosomes* and *chloroplast* have those enzymes which involved in protein synthesis and photosynthesis respectively.



Q.2 Define Catalysis, Substrate, Products.

Ans. **Catalysis** The process by which a chemical reaction is enhanced by a biocatalyst or enzyme without its own involvement in the product.

Substrate The molecule or molecules on which an enzyme exerts its catalytic action.

Products The formations occurred by the chemical reactions as a result are called products.

Q.3 Define activator and prosthetic group.

Ans. **Activator** The inorganic part of an enzyme is called activator. These are Mg^{++}, Fe^{++}, Zn^{++} and Cu^{++} etc.

It is that substance which makes another substance active. (See coenzyme for prosthetic group).

Q.4 Brief about cofactors and coenzymes.

Ans. **COFACTOR** The non-protein component of some enzymes that is necessary for catalytic activity which may be metal ions or coenzymes: (i) activators (ii) prosthetic group (iii) co-enzymes.

Function of Cofactor:

Cofactors play the role of bridge between enzymes and substrate. They provide a source of chemical energy. Mg^{2+}, Fe^{2+}, Zn^{2+} etc are used as cofactor. The detachable cofactor is called as an activator if it is an inorganic ion.

COENZYME An organic molecule i.e. vitamin that acts as an enzyme cofactor.

Example: NAD, NADP, Biotin and coenzyme A etc

Function of Coenzyme:

Coenzymes function as intermediate carrier molecules, which transfer or remove functional groups, atoms or electrons. When coenzyme is tightly bound to enzyme, in this case it is known as ***Prosthetic Group***. If it may be only loosely arranged then acts like a secondary substrate of the enzyme.

Q.5 What do you know about apoenzyme and Haloenzyme?

Ans. **Apoenzyme** The inactive protein portion of an enzyme that remains when the prosthetic group or cofactor has been removed.

OR

"The remaining protein of an enzyme after removal of cofactor or prosthetic group is called aponzyme". (Kinases requires Mg and Mn ions).

Holoenzyme The active complex, which has *enzyme plus cofactor*. In other words, sum of enzyme and cofactor is holoenzyme. Thus apoenzyme may become active by addition of cofactors.

Q.6 Where are enzymes found commonly?

Ans. **Sites of Enzymes**

Enzymes are present in *cytoplasm* and also found near the site of productions. They are also tightly bounded to cell organelles.

Respiratory enzymes are present in *Mitochondria*. *Chloroplasts* have that enzyme which is involved in photosynthesis. Enzymes help in protein synthesis are found in *ribosomes*.

All enzymes are produced within cells. Some enzymes speed up the reactions inside the own cell while some are produced inside its cells but play its role out side the cells, in a particular environment. On these basis enzymes are divided into two categories Endoenzymes and Exoenzymes.

FOR CONCEPT

Q. Define exoenzymes and endoenzymes.

Exoenzyme:

"Enzymes which come out of cell and catalyze the useful reactions in their environment are known as exoenzymes".

Example: Pepsin is secreted by chief cells and works in stomach's environment.

Endoenzymes:

"Enzymes which are formed within a cell and also act within the same cell is called endoenzymes".

Q.7 What is the difference between enzyme and proenzyme?

Ans. Enzymes like pepsin are not produced in such form, these are produced in inactive form i.e. pepsinogen. The inactive form of enzyme is called *Proenzyme*. This proenzyme is changed into active form by HCl.

$$\underset{\text{(Proenzyme)}}{\text{Pepsinogen}} \xrightarrow{\text{HCl}} \underset{\text{(Enzyme)}}{\text{Pepsin}}$$

Q.8 Write down the characteristics of enzymes.

Ans. **CHARACTERISTICS OF ENZYMES**

(1) **Globular Proteins:** All enzymes are globular proteins.

(2) **Speed up the Reactions:** Enzymes speed up or enhance the chemical reaction without being used up.

(3) **No Effect on End Product:** When the reactant or substrate change into product the enzyme does not effect on the nature of end product.

(4) **Minute Amount:** Very minute amount is very much effective for reactions.

(5) **Specific in Action:** A particular enzyme acts on a specific substrate or a special group of substrates.

(6) **Sensitive to Temperature:** All enzymes perform its function at a specific temperature called as ***optimum temperature***. Human enzymes have 37°C optimum temperature.

(7) **Sensitive to pH:** Different enzymes properly work at its optimum pH. e.g. Pepsin works on 2.00 pH and catalase on 7.60 pH.

(8) **Sensitive to Substrate Concentration:** Up to a certain limit, the rate of enzyme reaction increases with increasing substrate concentration.

(9) **Requirement of Co-factors:** Co-factors are essential for proper performance.

(10) **Lower the Activation Energy:** Enzymes lower the activation energy for reactions.

(11) **Enzyme Larger than Substrate:** Most enzymes are larger than substrates.

Some enzymes are potentially damaging if they are manufactured in their active form. For example, **pepsin** is a powerful protein-digesting enzyme and is quite capable of destroying cell's internal structure and thus is produced in inactive **pepsinogen** form by the cell. It is converted in its active form only in the digestive tract where it is required to be active.

Q.9 Write an account on the mechanism of enzymes or catalysis.

Ans. **MECHANISM OF ENZYME ACTION OR CATALYSIS**

(i) Lock and Key Hypothesis:

Enzymes are specific, they have particular shape. ***Three-dimensional globular proteins*** give specific structure and specific chemical composition. Actually, enzyme has a particular shape into which the substrate fit exactly. This is lock and key hypothesis.

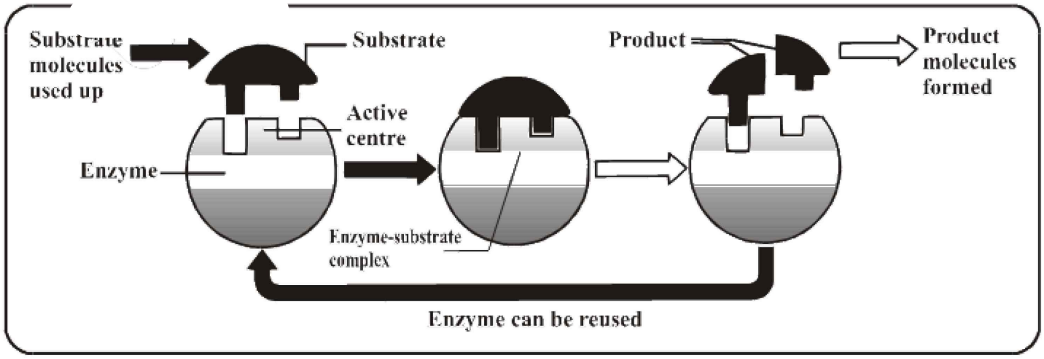


Fig. Diagrammatic representation of an enzyme-substrate reaction (Lock and Key Model)

(ii) Enzyme not Changed During Reaction:

Enzyme reacts with substrate and changes the substrate into products but enzyme itself not changed. It remains unaltered. After the formation of products, it goes toward a new reaction and again performs its function.

E + S----------(ES)----------Product + E

Enzyme Substrate Enzyme Substrate Complex (E ready for new work)

(iii) Active Site:

The site where the substrate binds in enzyme is known as active site.

Active site has specific shape. The enzyme is larger than substrate. The active site of the enzyme is smaller than its other body bulk. Only three 3 to twelve 12 amino acids may be arranged in active site portion. These amino acids have specific coiling and folding which becomes the reason of special symmetry or geometry.

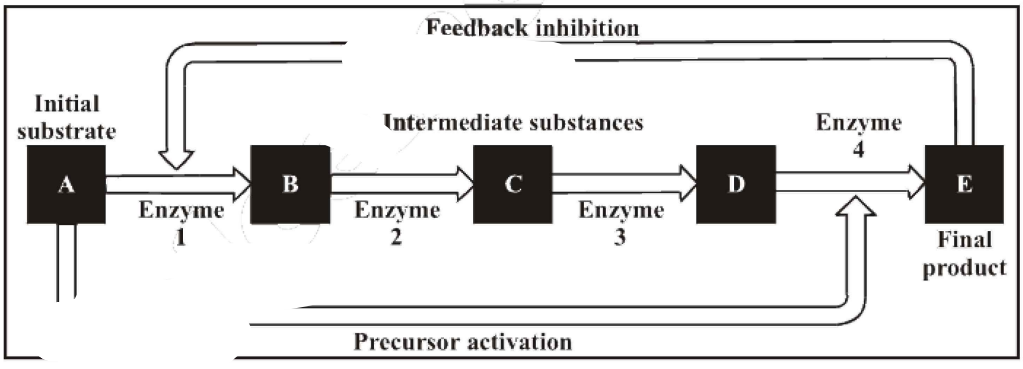


Fig. Enzyme to enzyme chain (association)

(iv) Binding and Catalytic Places of Active Site:

Active site of enzyme is divided into two further sites. One is binding site and other catalytic site.

Binding site holds proper substrate and fit it as ES-complex.

Catalytic site transforms the substrate into product or products.

(v) "Induced Fit" Hypothesis:

According to ***E. Fischer*** (1890), there is no modification or flexibility in the active site so lock and key rule is applied here. But in 1959 another hypothesis came about the active site. ***Mr. Koshland*** proposed that active site could be modified as the substrate interacts with the enzyme.

In other words, the slightly change may be occurred in active site and enables the enzyme to perform its catalytic activity more effectively.

EXAMINE YOURSELF

Q. Define Binding site, catalytic site, active site, apoenzyme, holoenzyme.

Q. What is "Lock and Key" hypothesis?

Q. Discuss induced fit hypothesis.

Q. What is enzyme substrate complex? Give an example.

Q.10 Which factors do affect the activity of enzymes?

Ans. **FACTORS AFFECTING THE RATE OF ENZYME ACTION**

Enzyme concentration, substrate concentration temperature and pH value are major factors, which affect the rate of enzyme action. Some factors should be constant and few should be at optimum level for proper rate of enzyme action. The work of an enzyme is disturbed by any factor which change the shape, chemistry or specificity of its.

(1) Enzyme Concentration:

The rate of reaction is proportional to enzyme concentration if temperature and pH remain constant. Increase the enzyme means number of active sites is increased. Substrate is converted into products actively due to available of extra active sites. These are all have a limit, after a certain limit there will be no increase in rate of reaction even increasing of enzymes.

(2) Substrate Concentration:

At constant enzyme concentration, the rate of an enzyme reaction increases with increasing substrate concentration. Ultimately, a point comes when any further increase in substrate concentration produces no significant change in reaction rate.

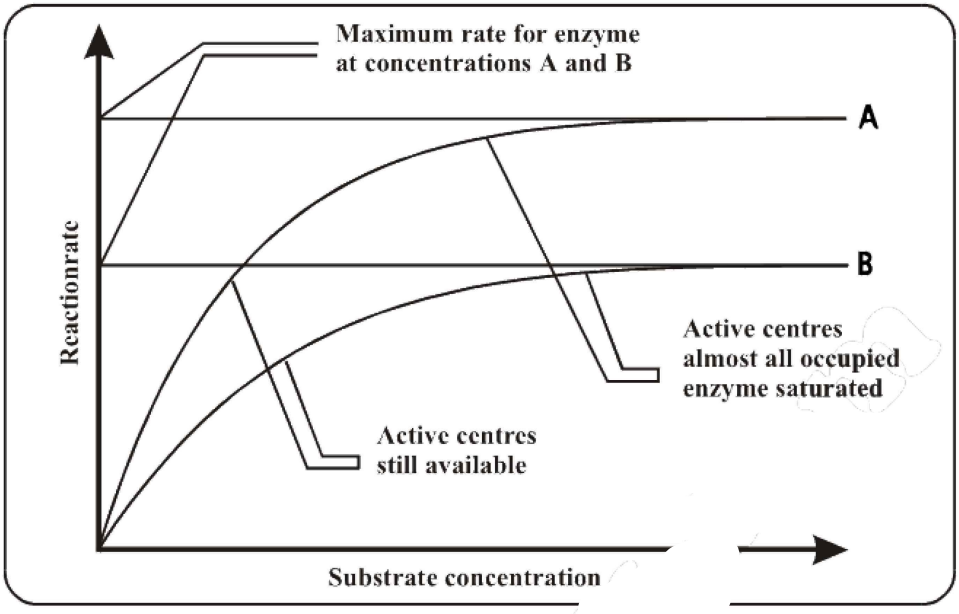


Fig. Effect of enzyme concentration and substrate concentration on the rate of an enzyme catalyzed reaction

(3) Temperature:

A suitable temperature is necessary for work of enzyme. An enzyme has maximum activity at a particular temperature this is optimum temperature. The enzymes of human body play a effective role at 37°C because it is optimum temperature for human enzymes.

In case of more than enough temperature, the rate of reaction is decreased. At this stage, the frequency of collisions is increased, in this way, the structure of an enzyme is disrupted and enzyme becomes denatured. In other word, due to extra collisions the structure of enzyme is disturbed and globular protein de-shaped thus inactive enzyme is called denatured.

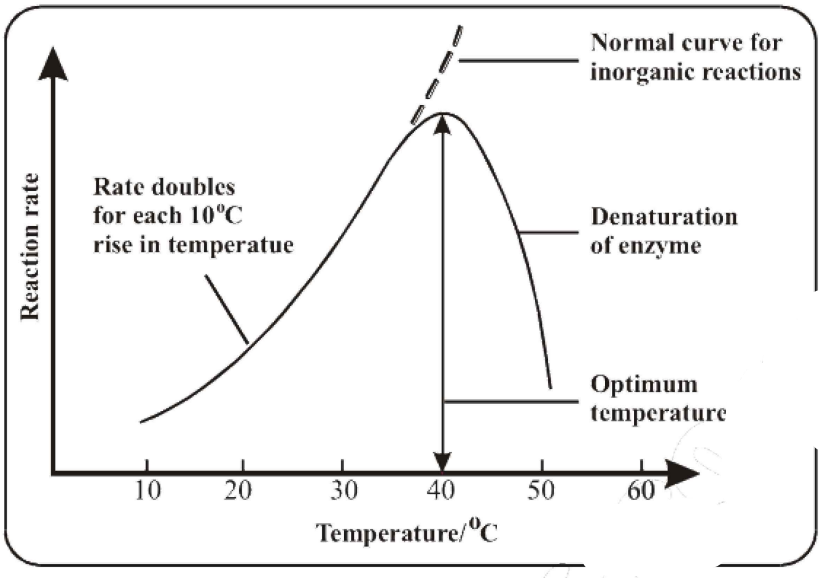


Fig. Effect of temperature on the rate of an enzyme catalyzed reaction

(4) Effect of pH Value:

Every enzyme functions efficiently over a particular pH range, at constant temperature. When the pH value is changed above or below the rate of enzyme activity diminishes.

Example: Pepsin catalyses protein at acidic medium i.e. pH 2.00 while trypsin also catalyses the protein but in alkaline medium i.e. pH.

pH change leads to an alteration of enzyme shape. Enzyme may become denatured in non-suitable pH. pH change also alters the ionization of amino acids.

Examples:

Enzymes	Optimum pH
Pepsin	2.00
Sucrase	4.50
Enterokinase	5.50
Salivary amylase	6.80
Catalase	7.60
Chymotrypsin	7.00 to 8.00
Pancreatic lipase	9.00
Arginase	9.70

Q. What type of role is played by enzyme concentration during catalysis? (Ans. See Q. 10 (1).

Q. What do you know about three kinds of co-factors? [Consult helpline of Q.1)

Q. Describe the effect of substrate cone during catalysis. [(Ans. See Q.10 (2)]

Q. Write the effect of temperature on enzyme action. [See Q.10 (3)]

Q. What is the effect of pH on enzyme activity? [See Q.10 (4)]

Q. What are optimum pH values of pepsin, sucrase, catalase and arginase? [(Consult Q.10 (table)]

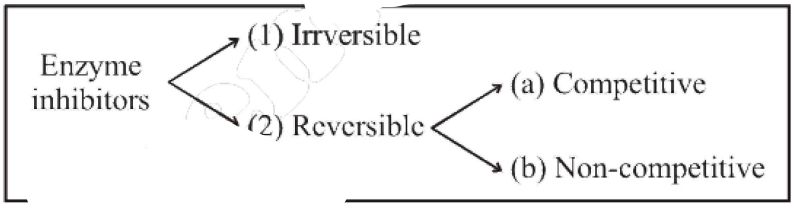
Q.11 Define Inhibitor, Inhibition and Explain Inhibitors.

Ans. **ENZYME INHIBITORS**

"The small molecules which fit into active sites or on the other parts of enzymes and reduce the rate of enzyme controlled reactions are called enzyme inhibitors".

Enzyme Inhibition:

"The reduction in rate of enzyme controlled reactions by poisons or drugs (chemicals) is called enzyme inhibition"



Explanation:

Active site is only for substrates. If active site of enzyme is occupied by other chemical like poison or drugs then the reaction rate is so much disturbed because of wrong thing on sensitive place. It is the reason of inhibition. And poisons or drugs are termed as inhibitors. Inhibitors are divided into two types:

(1) Irreversible Inhibitors

(2) Reversible Inhibitors

(1) Irreversible Inhibitors:

Those inhibitors which check the rate of reaction by occupying the active sites and destroying the structure of globulin (or protein or amino acids) are called irreversible inhibitors. I.I. form covalent bonds on active sites and block the site.

(2) Reversible Inhibitors:

In this case, weak linkages are found between enzyme and inhibitor. So, reversible inhibitors can be neutralized by increase of substrate concentration.

(a) Competitive Inhibitors:

Some times a compound has same structure to that of the normal substrate. This similar compound fits at active site. In this way, the active site owner i.e. substrate loose its site. Even a inhibitor after fitting no increasing the rate but place is occupied. Due to this reason a competition is occurred between inhibitor and substrate, so inhibitor is called competitive inhibitor.

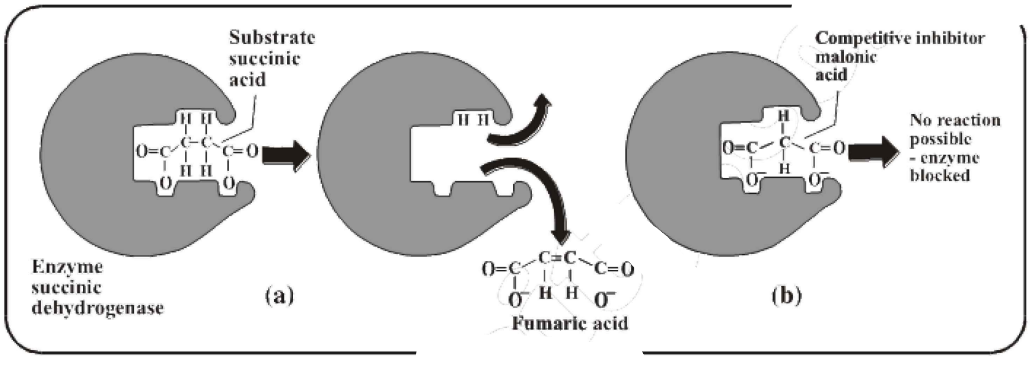


Fig. Mechanism of competitive inhibition. (a) Formation of enzyme-substrate complex resulting in the formation of product. (b) Inhibitor malonic acid does not fit the active site, hence no product is formed.

(b) Non-competitive Inhibitors:

In this type, inhibitor has no structural similarity to the substrate. So inhibitor does not fit at active site but attach to other part of enzyme. In this case substrate fits its own active site even then reaction rate is not increased.

DIFFICULT WORD MEANINGS

Words	Meanings	Words	Meanings
Enzyme	نامیاتی کیمیکل جو زندہ اشیاء کے ایکشن تیز کرنے	Complex	پیچیدہ/مرکب/بڑا
Active	متحرک	Lock and key role	انزائم اور سبسٹریٹ کا مخصوص تعلق
Specific	مخصوص	Proposed	بیان کردہ
Proceed	چلانا	Visualize	دکھائی دینا/اظہار
Coiled	بل کھاتا ہوا	Modification	بدلنا/تبدیلی
Substrate	ایسا کیمیکل جس پر انزائم عمل کرے	Concenteration	مقدار
Active site	انزائم کی وہ جگہ جہاں پر substrate سیٹ ہو	Available	پایا جانا/موجود
Contribute	حصہ ڈالے	Activity	سرگرمی/عمل/کام
Bridge	پل	Vibration	تھرتھراہٹ
Activator	متحرک کرنے والا	Denatured	اصل ساخت خراب ہونا/فطرت بدلنا
Essential	ضرور	Cause	وجہ
Raw material	خام مال	Narrow range	کم مقدار
Represent	اظہار	Antimetabolite	جو کیمیائی عملی کرنے والوں کے خلاف ہو
Consist of	پر مشتمل ہونا	Occupy	قبضہ کرنا/جگہ گھیرنا
Dissolved	حل ہوا	Drug	دوائی/میڈیسن
Globular	گول	Genuine	اصلی/خالص
Sensitive	حساس	Catalysis	ایکشن کو تیز کرنے کا عمل
Recognize	شناخت/نمائندگی	Inhibition	رکاوٹ
Contain	میں ہونا/میں رکھنا/میں پایا جانا	Inhibitor	رکاوٹ ڈالنے والا
Definite	مستقل	pH	تیزابیت یا اساسیت کے متعلق
Binding	بندھے ہونا	Optimum	مناسب مقدار

Q.1 Fill in the blank:

(i) Enzymes are composed of hundreds of ————————.

(ii) If the non-protein part is covalent bonded it is known as ————————.

(iii) Many enzymes require non-protein component called ———————— for their proper functioning.

(iv) Enzyme are highly ———————— in nature.

(v) The enzymes which carry out the synthesis of ———————— are integral parts of ribosomes.

ANSWERS:

(i) Amino acids (ii) Prosthetic group (iii) Co-factor

(iv) Specific (v) Protein

Q.2 Write whether the statement is 'true' or 'false' and write the correct statement if it is false:

	STATEMENT	T/F	CORRECT STATEMENT
(i)	Ligases catalyze the breakdown of complex substances into simple ones but water is not used as in hydrolytic reactions.	F	Oxidases catalyse the break of complex substances into simple ones but the water is used in hydrolytic reactions.
(ii)	Oxidases catalyze the transfer of hydrogen atoms to oxygen.	T	
(iii)	Calvin Malvin proposed Lock and Key model for enzyme action.	F	Emil Fischer proposed lock and key model for enzyme action.
(iv)	The active site of an enzyme is composed of four regions.	F	Active site of an enzyme is composed of one region or two regions.
(v)	Structure of an enzyme has no specific importance.	F	Structure of an enzyme has specific importance.

Q.3 Each question has four options. Encircle the correct answer:

(i) If more substrate to an already occurring enzymatic reaction is added, more enzyme activity is seen because:

(a) Then is probably more substrate present than there is enzyme.

(b) Then is probably more enzyme available than there is substrate.

(c) Then is probably more product present than their in either substrate or enzyme.

(d) The enzyme substrate complex is probably failing to form during the reaction.

(ii) What if you add more substrate to already occurring enzymatic reaction and it has no effect on the rate of reaction? What is the form given for this situation?

(a) Saturation (b) Denaturation

(c) Composition (d) Inhibition

(iii) The rate of enzyme – catalyzed reaction:

(a) Is constant under all conditions

(b) Decrease as substrate concentration increase

(c) Cannot be measured

(d) Can be reduced by inhibitors

(iv) The active site of an enzyme.

(a) Never changes

(b) Forms no chemical bond with substrate

(c) Determines, by its structure, the specificity of the enzyme

(d) Looks like a lump projecting from the surface of an enzyme

(v) Which statement about enzyme is not true?

(a) They consist of proteins, with or without a non-protein part

(b) They change the rate of catalyzed reaction

(c) They are sensitive to heat

(d) They are non-specific in their action

ANSWERS:

(i) (b) (ii) (a) (iii) (d) (iv) (c) (v) (d)

Q.4 Short Questions:

(i) List two conditions that destroy enzymatic activity by disrupting bonds between the atoms in an enzyme.

Ans. (a) Temperature (b) pH value

(ii) How do low and high temperature, respectively effect an enzyme activity?

Ans. Very high and low temperature reduce the activity of enzyme, Enzyme activity is at its best at optimum temperature.

(iii) What is a prosthetic group?

Ans. The non-portenious part attached with enzyme by covalent bond is called prosthetic group.

(iv) Define inhibitors of enzyme.

Ans. Inhibitors are chemical substance which block the active site of enzyme.

(v) How does an enzyme accelerate a metabolic reaction?

Ans. An enzyme accelerates a metabolic reaction because:

(a) It acts as a catalyst.

(b) It remains unchanged and can at in a reaction again and again.

(c) It works at very fast speed

(vi) What is the importance of enzymes in life?

Ans. Importance of enzymes [illegible]:

(a) It controls all the metabolic activities in a cell like respiration, photosynthesis.

(b) Enzymes being about synthesis of all complex molecules of life such as nucleic acid, protein, starch, glycogen, lipid etc.

(c) Each and every particular reaction in a cell is controlled by particular enzyme. If there is no enzyme, the reactions are slow down.