ENZYME

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Enzymes are biological catalyst catalyzing different biochemical reaction in the living system.

Enzymes are proteinaceous in nature.they exist in colloidal form.

Form precipitate with salt solution.

Action of enzyme affected by temperature and pH.

They specificity towards substrate and chemical reaction.

Enzymes required in small amount.

It remains unaltered at the end of thechemical reaction.

They are found in mitochondria(enzmes of citric acid cycle)

In cytoplasm(enzymes of glycolitic pathway)

In cell membrane(carrier of nutrient)

Substrate:these are the substance on which enzymes act.by the action of enzyme it is converted to product and the enzyme released in unchanged form.

Enzyme + substrate = product + enzyme

Enzymes are named by addition of suffix “ase” at the end of the substrate

Eg. Lipid hydrolysed by enzyme lipase(substrate-lipid)

Urea hydrolysed by urease(substrate-urea)

Enzymes are classified into six types OTHLIL

O-Oxidoreductase(catalyse oxidation and reduction of substrate.eg alcohol dehydrogenase,lactate dehydrogenase,glucose 6-phospahate dehydrogenase

Ethyl alcohol + NAD^+=ethanal +NADH+H^+

T-Transferase:catalyse transfer of a group from one substrate to anothrer substrate.eg. Transaminase,transcyclase,transpeptidase.

H-Hydrolases:catalyze hydrolysis of substrate.eg. Esterases,proteoses

L-Lyase:catalyze addition or removal of group from the substrate other than oxidation,reduction and hydrolysis. Eg. Carboxylase,decarboxylase,aldolase,enolase.

I-Isomerase:convert one compound into another. Eg.isomerase.epimerase.racemase

L-Ligase:catalyse the linking of two compounds and breakdown the compound.eg. DNA Ligase,Acetyl-CoA ligase.

Enzymes are specific in its action..their specificity classified into 3 types :i) Absolute specificity ii)group specificity iii)optical specificity

When enzymes catalyze only a particular reaction,it is called absolute specificity. Eg.urease hydrolyze urea

When enzyme acts on specific bond or linkage which is common to all substances,eg. Amylase enzyme acts on glycosidic linkage present in starch,dextrin and glucose

When enzyme acts on a particular type of optical isomer of the substance, eg. L-amino oxidase acts on L-amino acid.

Note:*proenzyme /zymogen-inactive form of enzyme.it is synthesized in inactive form which later converted to active form*.

Eg. Pepsinogen (inacive) → pepsin (active)

Isoenzyme-these are the enzymes obtained from different sources have different physical and chemical properties but catalyze the same reaction.

Lysozyme:this is an enzyme present in human tears,saliva,gastric juice hydrolyses acetyl amino polysaccharides of bacterial cell wall..thus protect bdy against bacteria…

Eg.Lactate dehydrogenase (LDH) exist in 5 forms LDH_1,LDH_2,LDH_3,$LDH_4$and LDH_5…these enzymes have different physical properties but have the same function i.e oxidation of lactic acid to pyuvic acid.

Factors affecting enzymatic action:

rate of enzymatic action directly propotional to temperature….upto 40c the rate of enzymatic actio increases but beyond this temperature rate of enzymatic action becomes *slow…40c called as optimum temperature.*

pH:with increase in pH rate of enzyme action increases till rate achieve the maximum…then start decreasing on further rise in Ph…this is called *optimum pH.*

Rate of enzyme action is directly propotional to enzyme concentration if sufficient substrate concentration is there to react with enzyme..if substrate concentration is less than enzyme concentration then on saturation there will be no effect.

Substrate concentration: is directly propotional to rate of enzyme action..upto a certain point after this increase in concentration of substrate has no effect in enzyme action.

Radition:α,β,γ ray,uv-ray,x-ray inacivate enzyme action by formation of peroxide which oxidise enzyme…

Activators:certain metal ions and co-enzymes activete enzymes.

Inhibitors:enzymes inacivated by inhibitors like silver and mercury and iodine.

ENZYME INHIBITION: Enzyme inhibitors are the substances which lower down the enzyme action.they produce their effect by acting on the co-enzyme,apoenzyme or prosthetic group.

Enzyme inhibition is classified as:i) competitive ii)noncompetitive iii)allosteric

Competitive:inhibitor and substrate both have similar strucure compete for the same enzyme…in this case inhibitor is more powerfull and forms "inhibitor –enzyme complex"…there is no formation of product…it is a reversible type…

Eg. Conversion of succinic acid to fumaric acid in presence od succinic acid dehydrogenase but malonic acid having same chemical strucure acts as competitive inhibitor.

Non-competitive inhibitor: here the inhibitor acts on enzyme substrate complex and prevents product formation,doesnot need structural similarity between substrate and inhibitor.

ES + I = ESI = NO PRODUCT

ALLOSTERIC INHIBITION : enzyme possess an allosteric site in its structure,inhibitor binds to allosteric site produces confirmational changes in the enzyme,sothat substrate cannot bind to the enzyme and product is not formed.

DIAGNOSIS OF DISEASE BY ENZYME:

There are certain diseases estimated by presence of certain enzyme in the blood.

List of some enzymes and diseases are given below

i)Amylase –Acute pancreatitiss

ii)Alkaline phospahatase –Rickets

iii)Creatinini pjophakinase –Myocardial infarction(MI)

iv)Glutamic oxaloacetic transaminase –MI

v)Glutamic pyruvic transaminase –Liver disease

vi)Isocitrate dehydrogenase –Hepatitis,Liver metastasis

vii)Lactate dehydrogenase(LDH) –MI

vii)Lipase –Acute pancreatitis

PHARMACEUTICAL AND THERAPEUTIC ACTION OF ENZYMES :

Therapeutic use of enzyme:

i)Enzymes pepsin,papain and amylase for improving digeston.

ii)Enzyme Hyaluronidase for improving diiffusion of drugs.

iii)Enzymes streptokinase and Urokinase for dissolving blood clot.

iv)Enzyme trypsin for treatment of catract.

v)Enzyme Asparginase for treatment of cancer.

Pharmaceutical application:

Enzymes often bind with the target with higher affinity and specificity and catalyze and convert multiple drug molecule to the desired product.

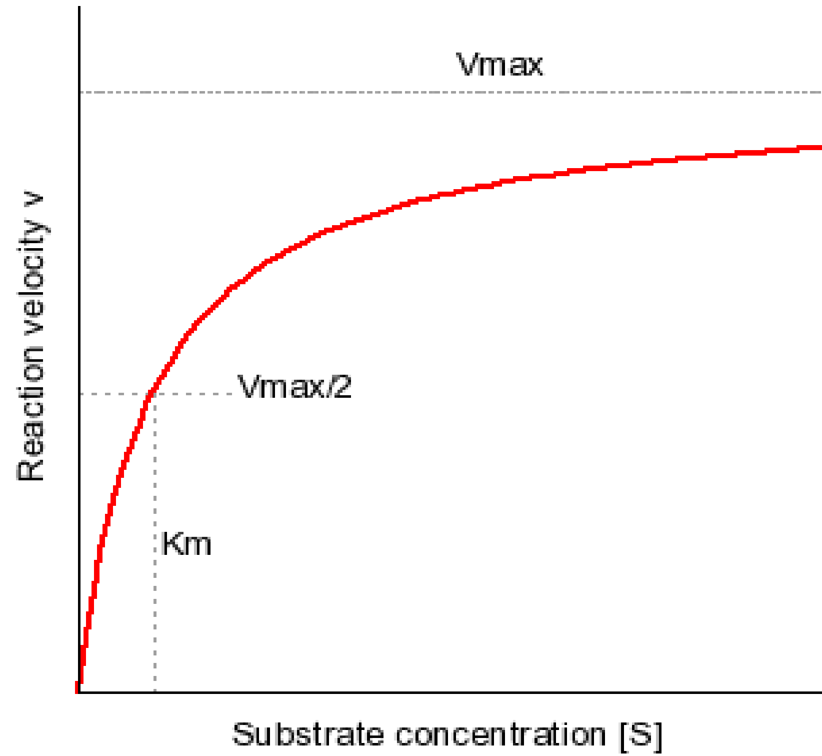
i)Enzyme penicillin acylase produce 6-aminopenicillanic acid from penicillin-G, 6-aminopenicillanic acid used in production of β-lactam antibiotics.

ii)The enzyme glucose oxidase is needed for production of fructose syrup.

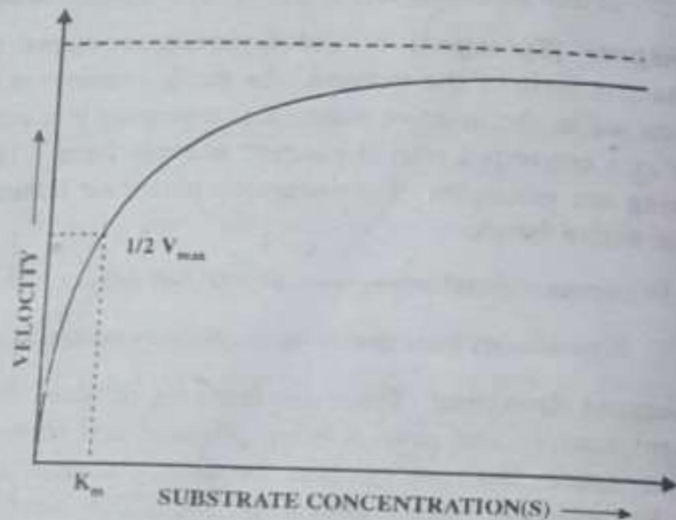
iii)Amulase is needed for the production of dextrin.

iv)The enzyme papain is needed for production of protein hydrolase.

ENZYME ACTION : MICHAELIS CURVE



V_{max} is reached when all the enzyme molecules are saturated with the substrate. So there is no increase in velocity. The curve is called *Michaelis curve*.



Effect of substrate concentration on the velocity of enzyme catalysed reaction

The mathematical equation is called *Michaelis Menton equation* which is as follows.

$$V_o = \frac{V_{max}[S]}{K_m + [S]}$$

Where V_o = Initial velocity

V_{max} = Maximal velocity

[S] = Substrate concentration.

K_m = Michaelis constant.

Km is equal to $\frac{1}{V}$. i.e. Km is equivalent to substrate concentration required to produce half maximal velocity.

Lineweaver - Burk plot : The drawbacks of Michaelis curve are:

1. Only an approximate but not an accurate value of Km can be obtained.
2. It is difficult to determine V_{max} accurately. It is because, V_{max} is only approached and never attained.
3. It is only a hyperbolic curve and not a straight line graph. So interpolation of data is not possible.

These drawbacks are overcome by a straight line graph called *Lineweaver - Burk plot*. It makes use of receprocals of Vo and [S] i.e. $\frac{1}{V_o}$ and $\frac{1}{[S]}$.

As we have already seen, Michaelis Menton equation is

$$V_o = \frac{V_{max}[S]}{K_m + [S]}$$

Reciprocal of this equation is

$$\frac{1}{V_o} = \frac{K_m + [S]}{V_{max}[S]}$$

It can be rewritten as

$$\frac{1}{V_o} = \frac{K_m}{V_{max}[S]} + \frac{[S]}{V_{max}[S]}$$

It can be simplified as

$$\frac{1}{V_o} = \frac{K_m}{V_{max}} \times \frac{1}{[S]} + \frac{1}{V_{max}}$$

The above equation is called as Lineweaver-Burk equation.

Now, a graph can be constructed by plotting $\frac{1}{V_o}$ on the y axis and $\frac{1}{[S]}$ on the X axis. This gives a straight line. From this graph three points are very clear and they can be easily determined :