19

Faculty of medicine – JU2018

◉ Sheet ○ Slides

DONE BY

Lujain &Lujain

CONTRIBUTED IN THE SCIENTIFIC CORRECTION

Lujain Akram

CONTRIBUTED IN THE GRAMMATICAL CORRECTION

Lujain Alaqtash

DOCTOR

Mamoun Ahram

-Classification of enzymes:

1- structure:

We said that enzymes can be simple or complex ➡ simple: all composed of **polypeptides**

Complex: can be conjugated with

Something that is not a protein.

The non-protein can be:

1-an organic compound .EX: Heme

2- metal, EX: Zn , Mg ,...

As a result we can call enzymes as :

- Holoenzymes :made of protein and non protein components .(active)
- Apoenzymes : (inactive) made of only proteins .

2- **Naming of enzymes :**

- in general enzymes end with the suffix (-ase)

-sometimes the name of the enzyme indicates exactly what this enzyme does

EX: Adenosine deaminase (removes an amino group from Adenosine) .

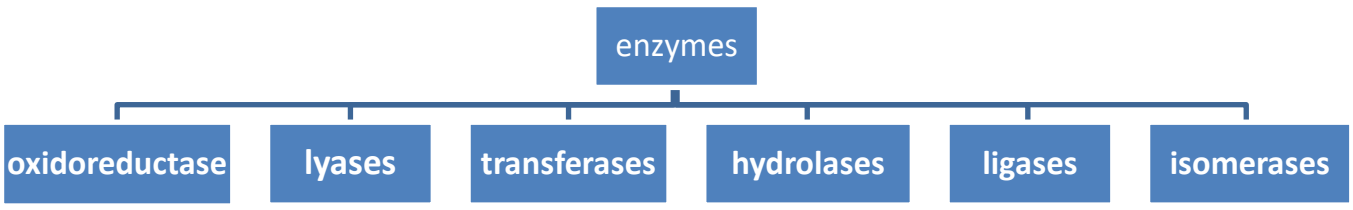
ATPase (breaks down ATP)

ATPsynthesase (synthesizes ATP)

- some enzymes have common names and we cannot tell what exactly the do by their names .

EX: Trypsin (a proteolytic enzyme = breaks down proteins into smaller peptides and amino acids).

3- Enzyme classes according to function :



1- Oxidoreductases :

This group is the largest and it's divided into 4 subgroups

a-dehydrogenases

b-oxidases

c-peroxidases

d-oxygenases

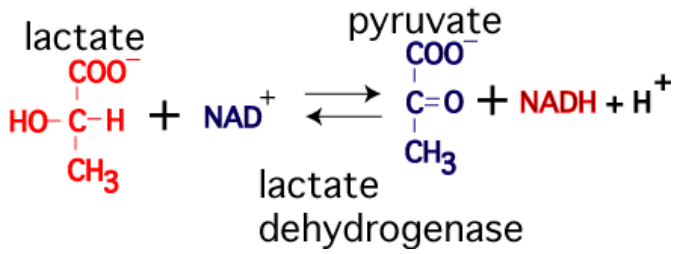
Function: they catalyze oxidation/reduction reactions involving the transfer of hydrogen.

1-a: Dehydrogenases :they help transfer electrons (either extraction or donation) in the form of hydride ions H- or hydrogen atoms using an electron-transferring coenzyme , such as NAD+, NADH or FADH2 .

Tip: how to recognize this kind of enzyme from any reaction ?

From the presence of NAD+/NADH OR FADH/FADH2.

EX: lactate dehydrogenase (important to know the reaction)

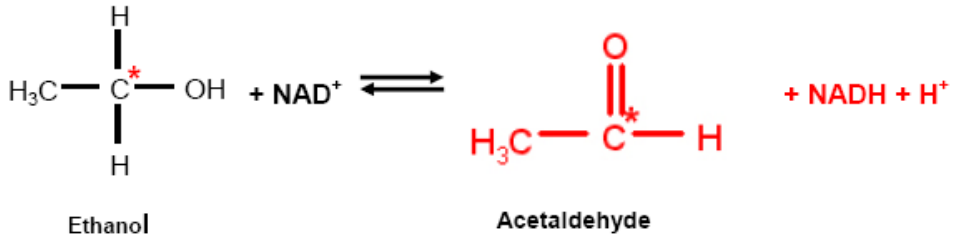


The function of lactate dehydrogenase is to oxidize lactate and convert it to pyruvate .

- The reverse reaction in which pyruvate is converted into lactate takes place in anaerobic glycolysis when there is no oxygen .

Note: the lactate dehydrogenase still catalyze both the reaction and the reverse

EX2: Alcohol dehydrogenase :



In yeast Pyruvate is converted into acetaldehyde and then to Ethanol.

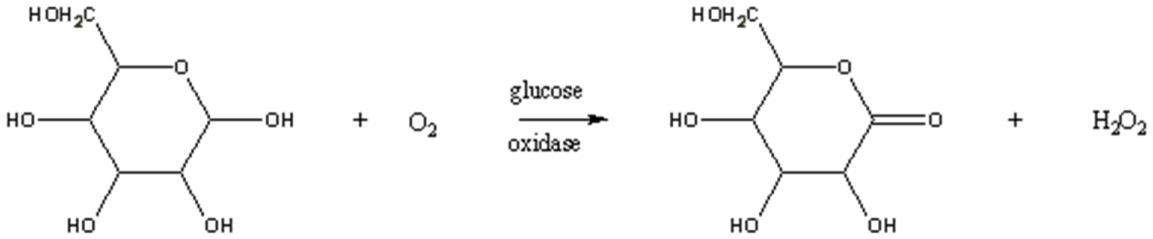
1-b: Oxidases :

They catalyze hydrogen transfer from the substrate to molecular oxygen producing hydrogen peroxide as a byproduct (H2O2)

Tip: presence of

- O2 in reactants -H2O2 in the products

EX: β-D-glucose + O_2 ⇆ gluconolactone + H_2O_2



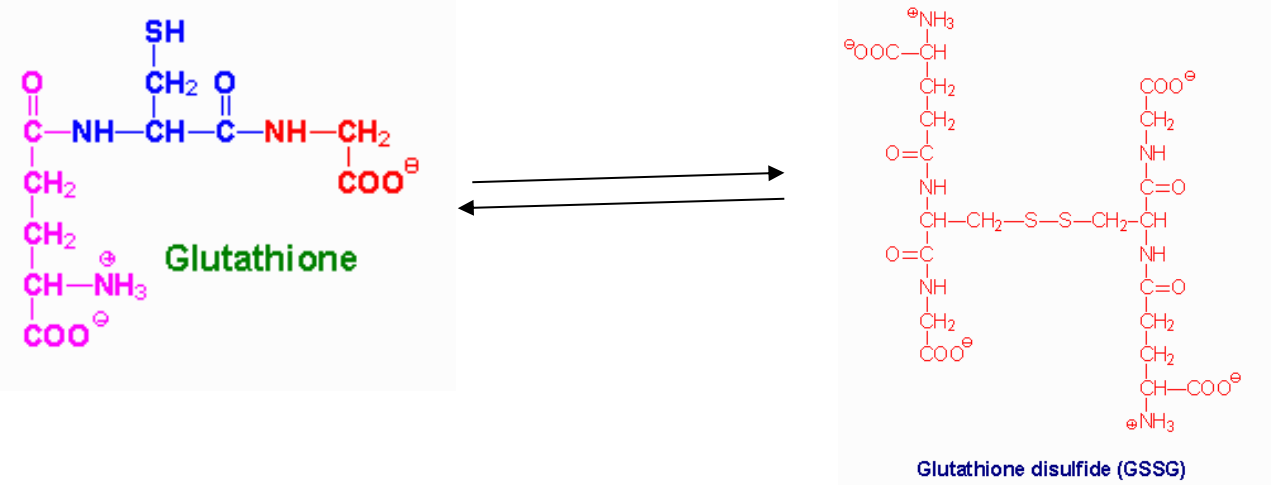
The enzyme is called glucose oxidase

1-c:peroxidases

They catalyze oxidation of a substrate by hydrogen peroxide , since H2O2 is harmful , it is removed by being reduce and the substrate is then oxidized.

Example: oxidation of two molecules of glutathione (GSH) in the presence of hydrogen peroxide:
The enzyme is glutathione peroxidase.

2 GSH + H_2O_2 ⇆ G-S-S-G + 2 H_2O



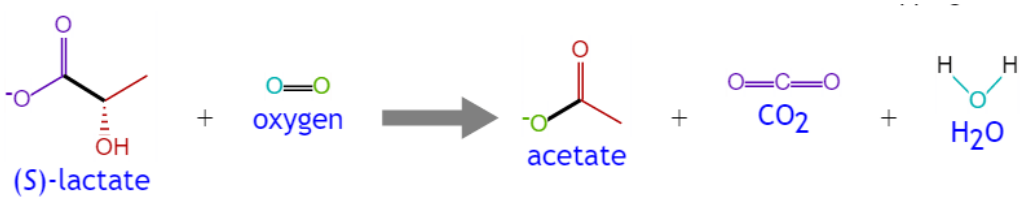
00.10.00

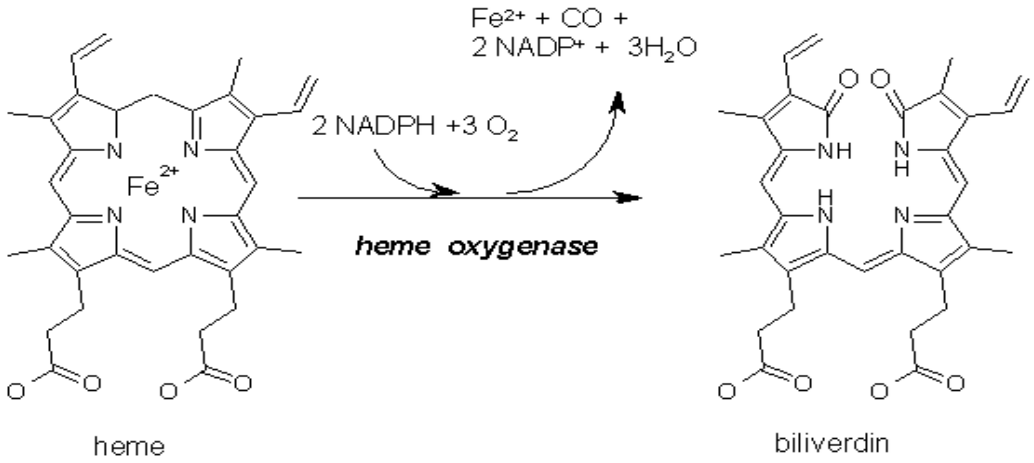
1-d:oxygenases

catalyze substrate oxidation by molecular oxygen through introducing O2 into the substrate (they transfer one oxygen atom to the substrate or 2 oxygen atoms) thus , the enzyme is either monoxygenase or dioxygenase .

Tip: O2 in reactants H2O in products (not H2O2)

Examples: lactate-2-monooxygenase and heme dioxygenase





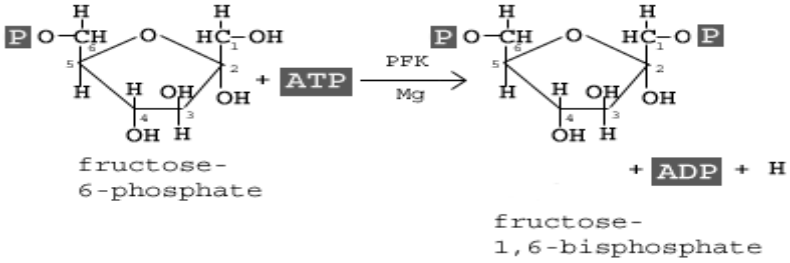
What is the difference between oxidase and oxygenase ??

They both have O2 in the reactants BUT, in oxidase you have hydrogen peroxide in the products while in oxygenase what happens is a transfer of oxygen to the substrate and this produces water .

2. Transferases: these enzymes transfer a functional group ex(C,N,P,S) from one substrate to an acceptor molecule .

a.An important type of transferases is Kinase (which are enzymes that phosphorylates other molecules) .

Phosphofructokinase : catalyzes transfer of phosphate from ATP to fructose-6-**phosphate**

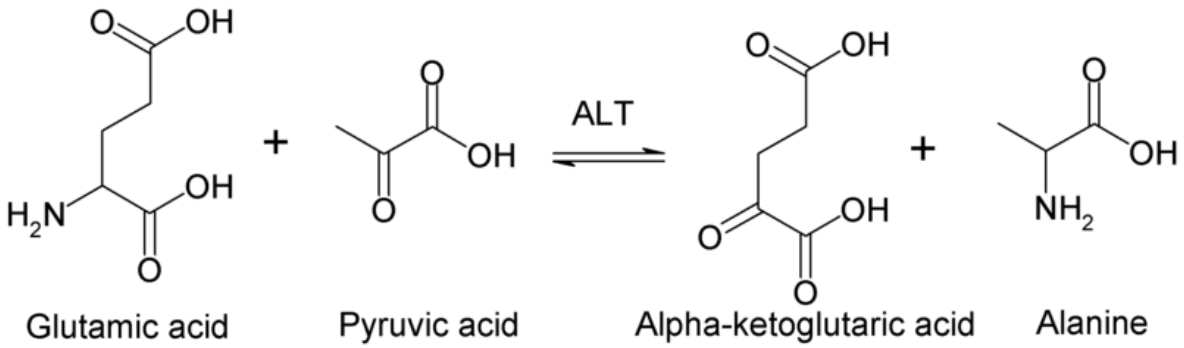


** Remember :metabolism of glucose : glucose is phosphorylated by hexokinase into glucose-6-phosphate which is then isomerized into fructose-6-phosphate and then its phosphorylated by phosphofructokinase into Fructose-1,6-bisphosphate .

b. another type of transferases is : Transaminases :

they transfer an amino group from an amino acid to a Keto acid ⟶ thus the amino acid becomes a keto acid and the keto acid becomes an amino acid . they also catalyze the inverse reaction . (Interconversion of amino acids)

so when the body needs a certain amino acid it can make it by a transaminase reaction.



* ALT=Alanine transaminase .(it's a very important enzyme , in laboratories when checking the lever's health they check for ALT enzyme) .

*alpha-ketoglutaric acid : its one of the intermediates of Krebs cycle , if you think about it , we can interconvert not only one amino acid to another , we can ALSO, use amino acids to produce energy through Krebs cycle .

3- Hydrolases :

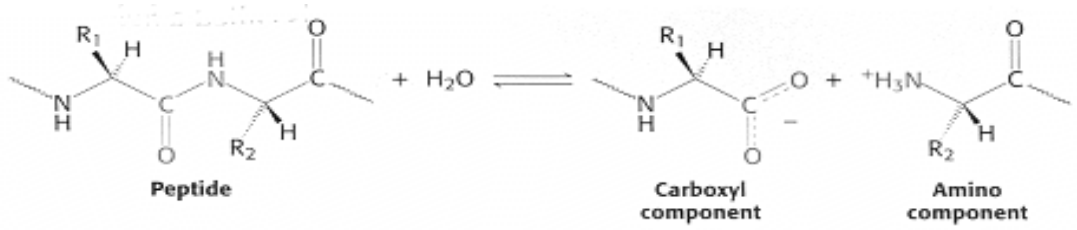
- They use water to break bonds, they catalyze cleavage reactions using water across the bond being broken or the fragment condensations.

EX: peptidases(they break the peptide bond),Esterases (break the ester bond) ,lipases (cleave lipids).... glycosidases , phosphatases are all examples of hydrolases named depending on the type of bond cleaved .

***note: phosphatases are the opposite of Kinases , kinases transfer a phosphate group from a molecule to another , to remove the added phosphate group, phosphatases function using water to remove the phosphate .**

Ex: to convert glucose-6-phosphate into glucose we use Glucose phosphatase .

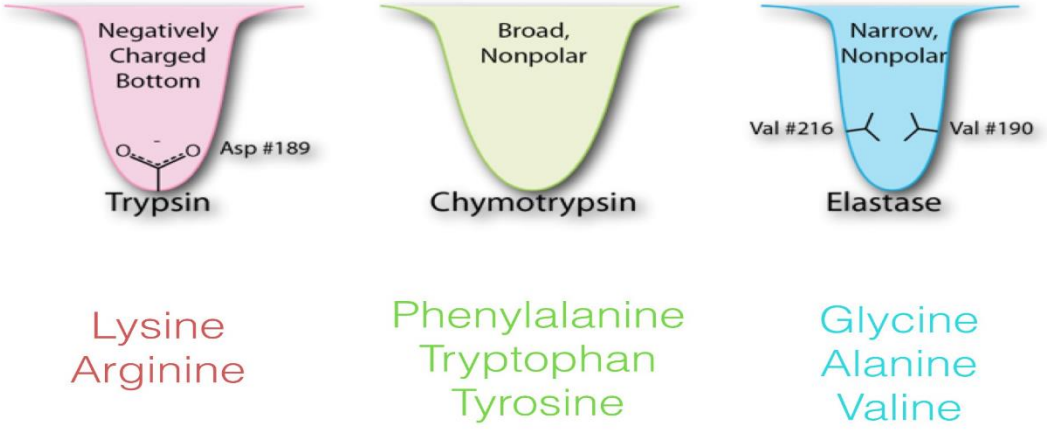
- A class of hydrolytic enzymes is proteases.



*These enzymes catalyze proteolysis (the hydrolysis of a peptide bond within proteins)

Note: proteases work on the 3 dimensional structure while peptidases work on peptides

- Trypsin breaks up peptide bonds only on the carboxyl side of Lys and Arg residues.
- Chymotrypsin hydrolyzes peptide bonds involving bulky aromatic amino acids.
- Elastase hydrolyzes peptide bonds involving small, uncharged groups such as Ala, Val, or Gly.

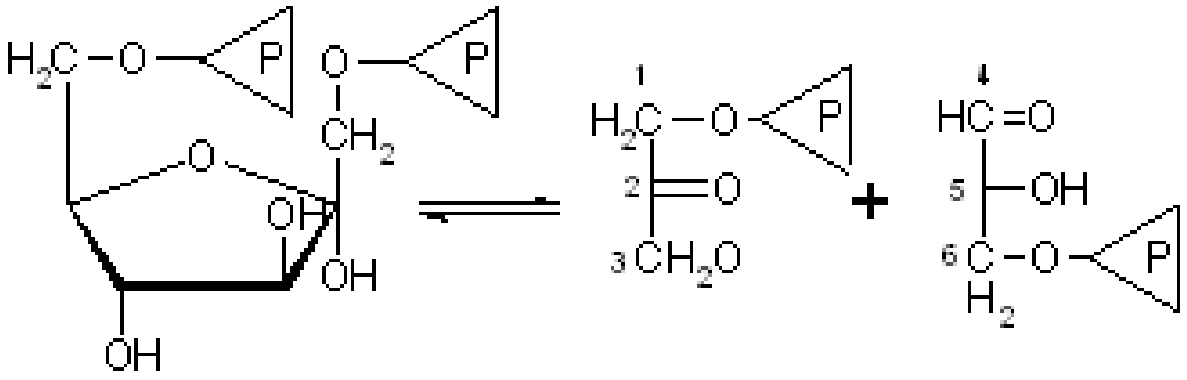


4-Lyases:

They can remove groups from substrates resulting in the formation OR removal of double bonds between C-C, C-O, C-N . without hydrolysis (elimination reaction)

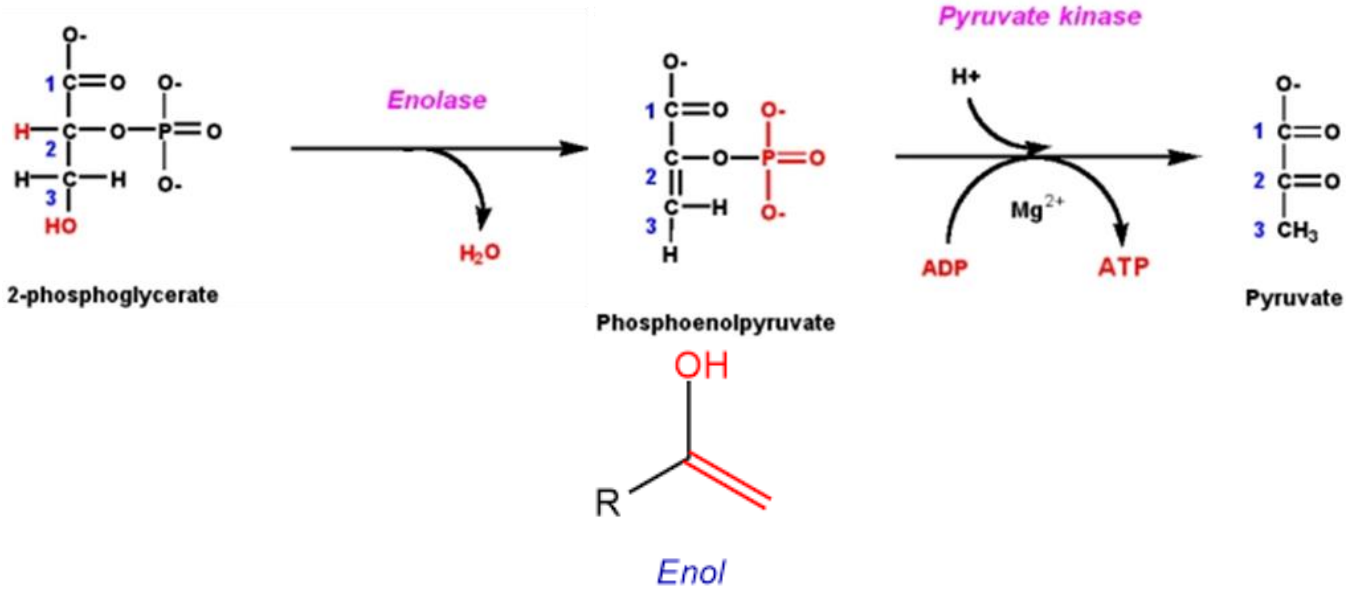
- These enzymes remove groups from substrates with the associated formation or removal of double bonds between C-C, C-O and C-N without hydrolysis (elimination reaction).
- Example: aldolase breaks down fructose-1,6-bisphosphate into dihydroxyacetone phosphate and glyceraldehydes-3-phosphate.

Fructose 1,6 bisphosphate ⇆ DHAP + GAP



Another example is Enolase , it's a lyase , it converts 2-phosphoglycerate to phosphoenolpyruvate an by formation of double bonds.

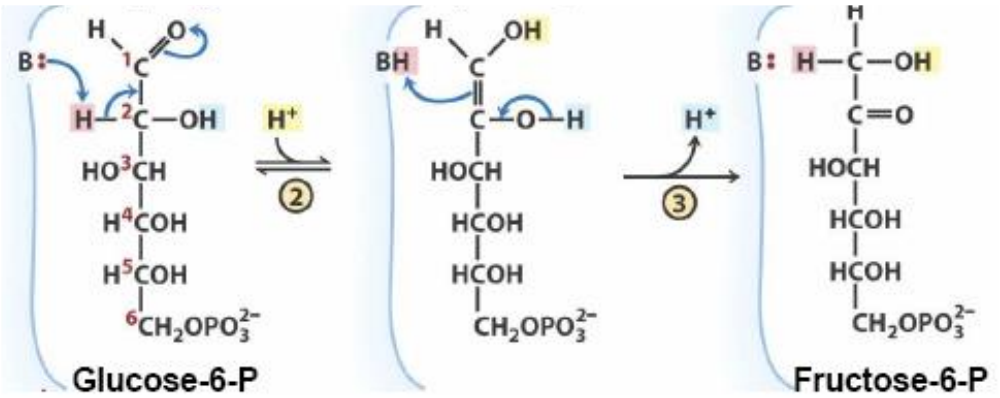
What it does is that it removes a water molecule by an elimination reaction resulting in the formation of an Enol molecule (formation of a double bond) then there is a pyruvate kinase that transfers a phosphate group from phosphoenolpyruvate to an ADP to eventually produce pyruvate



Enol

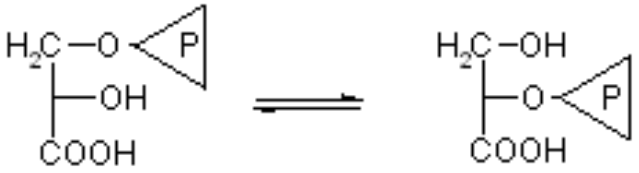
5- isomerases :

These enzymes catalyze intramolecular rearrangements.
Phosphoglucoisomerase isomerizes glucose-6-phosphate to fructose-6-phosphate.



Another EX is phosphoglycerate mutase which transfers a phosphate group from carbon number 3 to carbon number 2 of phosphorylated glycerate

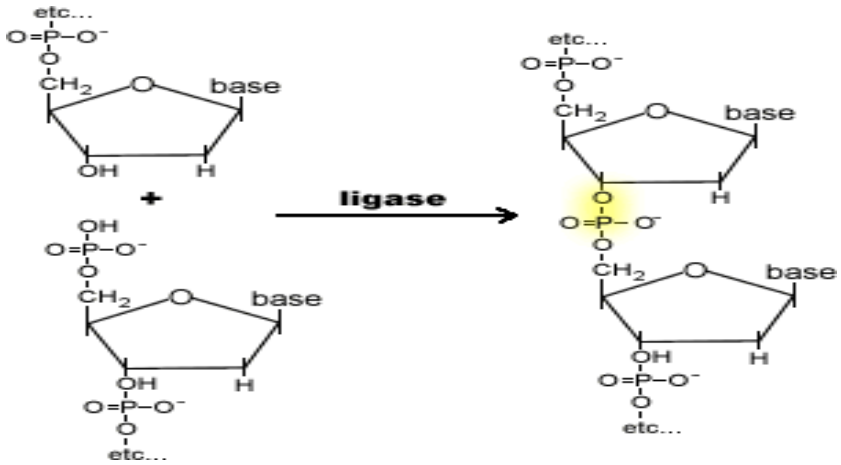
3-P glycerate ⇆ 2- P glycerate



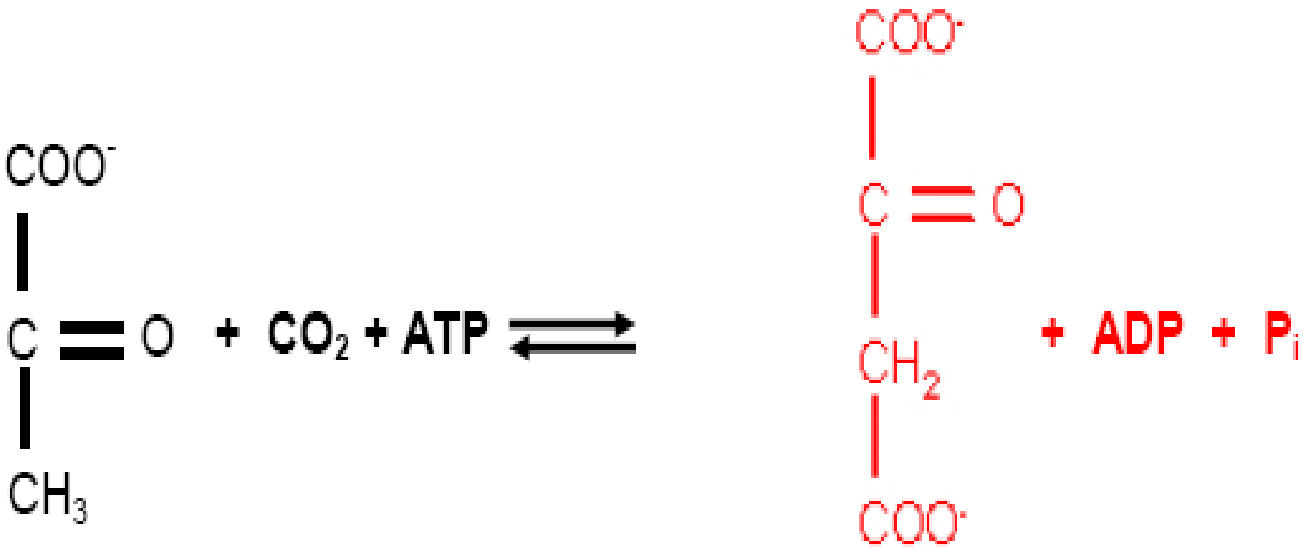
The examples used are almost all from the glycolytic pathway (the metabolism of glucose)

6-Ligases:

- Ligases join C-C, C-O, C-N, C-S and C-halogen bonds.
- The reaction is usually accompanied by the consumption of a high energy compound such as ATP and other nucleoside triphosphates.
- The ATP is involved in the transferases and ligases catalase reactions , the difference is in the transferases the phosphate is taken from ATP and it is add to the substrate ,in ligases we have ATP involved in the reaction but as a product we have a relase of inorganic phosphate and ADP .



- Example: pyruvate carboxylase:
- pyruvate + HCO_3^- + ATP ⇆ Oxaloacetate + ADP + Pi

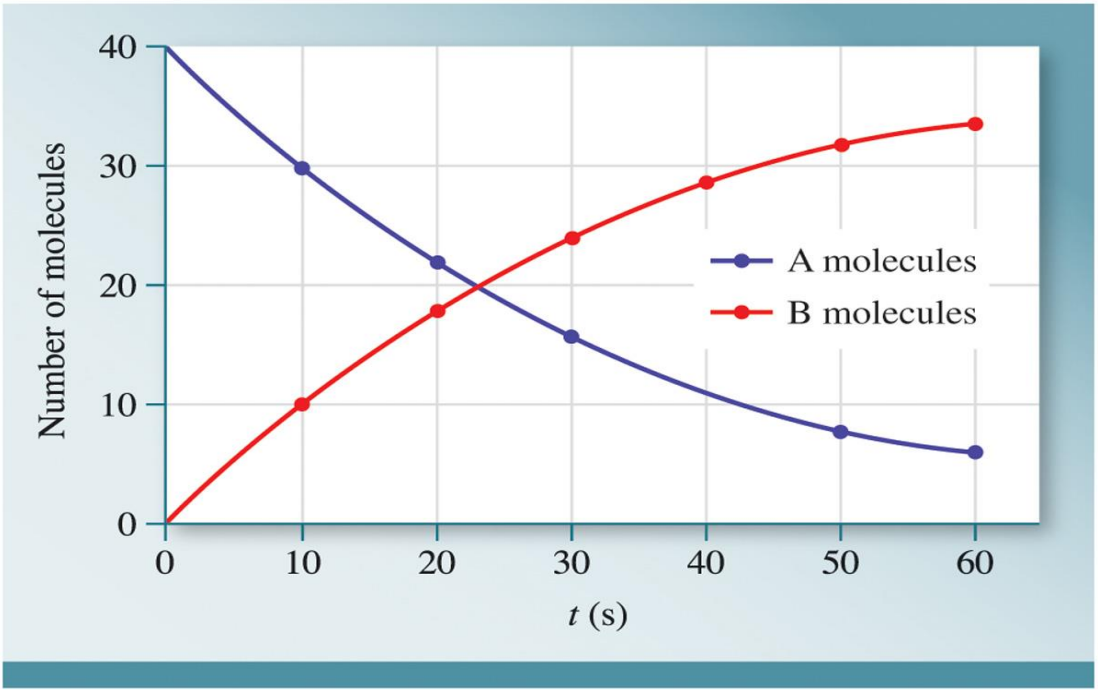


Enzymes ||

- Kinetics deals with the rates of chemical reactions.
- Chemical kinetics is the study of the rates of chemical reactions.
- For the reaction (A → B), velocity (*v)* or rate of reaction is the amount of B formed or the amount of A consumed per unit time, *t*. That is,

- Rate of reaction (velocity or v) = $-\frac{\Delta [A]}{\Delta t}$ *or* $\frac{\Delta [B]}{\Delta t}$

- In another words it is the rate of disappearance of the substrate or the rate of appearance of the product.



Rate law: The relationship between reaction rate and concentration of reactant(s) , which describes *how concentrations of reactants affect the rate of the reaction during a certain period*.

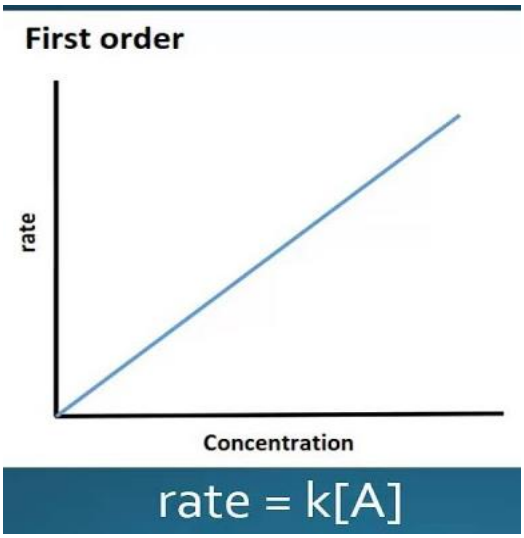
- For the reaction (A → B), the rate law is :

$$v = \frac{-d[A]}{dt} = k[A]$$

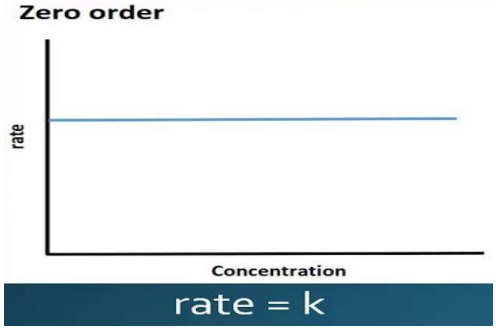
- Note: the rate is directly proportional to the concentration of A, and *k* which is the rate constant.
- *k* has the units of (time)$^{-1}$, usually sec^{-1}.
- If the concentration is doubled, the rate will also be doubled.

- If there is one substrate and this substrate is the only factor that affects the rate of the reaction we call this a **first order reaction** .**Rate =k{A}**

The plot indicates that the rate of a reaction increases linearly with increasing substrate concentration.

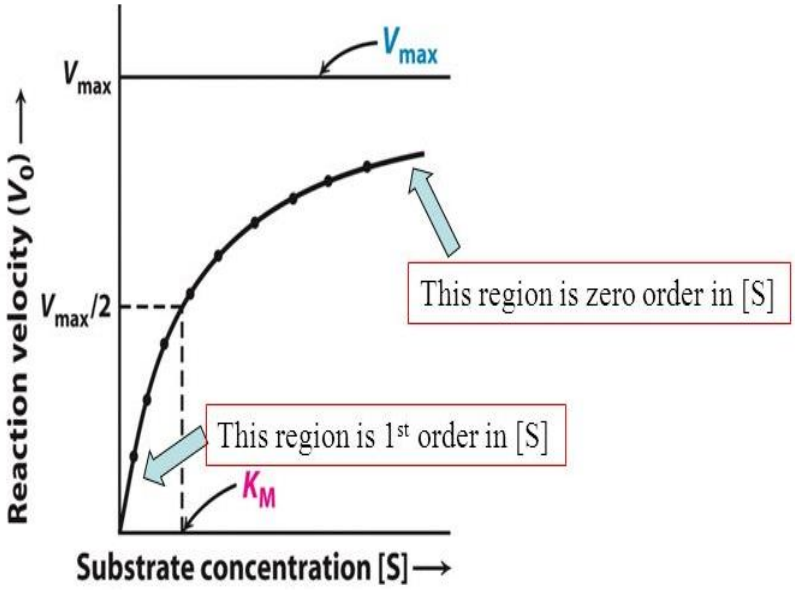


If the rate of the reaction is independent on the concentration of the substrate wecall this a **Zero order reaction.** rate = k$[A]^0$ = *k*



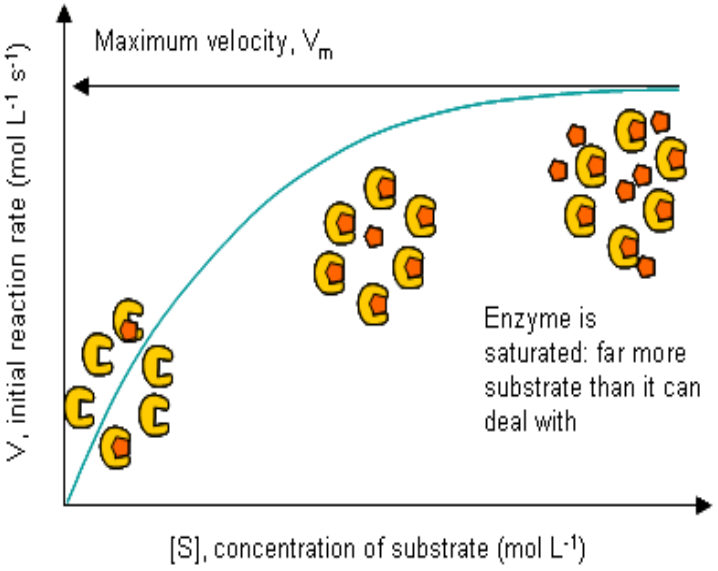
- Enzyme kinetics: studying the biological roles of enzymatic reactions.
- Enzyme-catalyzed reactions have hyperbolic plots.

Initial velocity (V_0) varies with the substrate concentration [S] where the rate of catalysis rises linearly as substrate concentration increases and then begins to level off and approach a maximum at higher substrate concentrations.



So the question is why dose it level off???
This is related to saturation which means at low cocentration of substrate some enzymes are free of substrate so you can add more substrate and you will have more enzymes get saturated , once you get to a very high cocentration of substrate now enzymes are all saturated and if you add a little more of substrate nothing will happen so the rate of the reaction is now constant and it is independent on the substrate concentration ,but it depends on the enzyme (how effecint the enzyme is and how much enzyme you have).

The hyperbolic plot is known as a saturation plot because the enzyme becomes" saturated" with substrate, i.e. each enzyme molecule has a substrate molecule associated



Further explanation: -At a fixed concentration of enzyme, V_o (initial velocity) is almost linearly proportional to [S] (substrate concentration). -However, V_o is nearly independent of [S] when [S] becomes larger.
-Vmax is achieved when the cataletic sites on the enzyme are saturated with substrate .

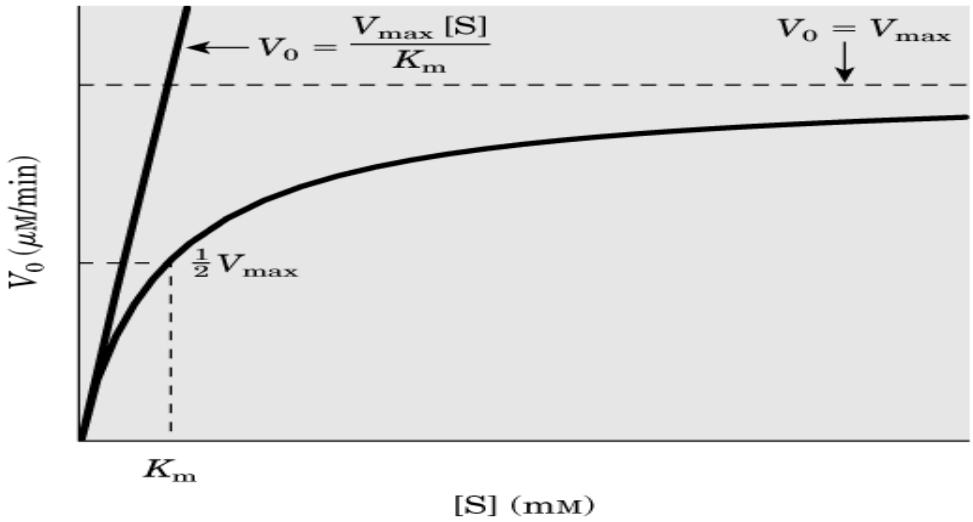
- Vmax reaveals the turnover number of an enzyme which is the number of substrate molecules converted into product per unit time when the enzyme is fully saturated. It is a measure of the catalytic ability of the enzyme.

The Michaelis-Menten Equation:

The Michaelis-Menten equation is a quantitative description of the relationship between the rate of an enzyme catalyzed reaction (V_0), substrate concentration [S], a rate constant (K_M) and maximal velocity (Vmax).

$$V_0 = V_{max} \frac{[S]}{[S] + K_M}$$

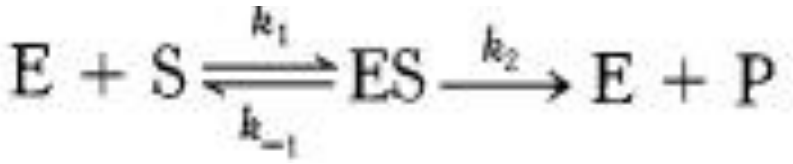
- **K_M (michaelis constant):** the substrate concentration when the velocity of the reaction is half of (Vmax) or when half of the active sites are filled



- K_M is the concentration of substrate at which half the active sites are filled.
- When [S] = K_M, then V_0 = Vmax/2
- Therefore, it provides a measure of enzyme affinity towards a substrate.
- The lower the K_M of an enzyme towards a substrate is, the higher its affinity to the same substrate is.this means you need low concentration of the substrate to get to half of **Vmax .**
- The lower the **[S]** the lower the velocity, but when the substrate concentration is very very high (much higher than **K_M)** the value of K_M become insignificant (**[S]** will be almost the same of **[S]+ K_M**)so V will be equal to Vmax .
  For more understanding let's say we have a substrate concentration of 0.1 and **K_M**=1 the velocity will be equal to Vmax *{ 0.1/(0.1+1)}=Vmax *0.09
  But if we have a substrate concentration of 100 and **K_M**=1 the velocity will be equal to Vmax *{100/(101)}= Vmax .

So at very low substrate concentration V_0 is really controlled by how much substrate there is , but when we have high substrate concentration V_0 become independent on how much substrate there is .

❖ For a reaction :



$$K_M = \frac{k_{-1} + k_2}{k_1}$$

$k_{-1} >> k_2$),
so $K_M = k_{-1}/k_1$

Each one of these arrows in the reaction has it's own k (constant) value.

- ES complex can dissociate because the interaction between the substrate and the enzyme is **reversible non covelent interaction**.
- K_M is related to the rate of dissociation of substrate from the enzyme to the enzyme-substrate complex (association).
- K_M *describes* the affinity of enzyme for the substrate, but is not an accurate measure of affinity , because we have **k_2** which is very small in value relative to **k_{-1} and k_1** .

K_D (dissociation constant) is the actual measure of the affinity..

$$K_D = (k\text{-}1/k1)$$

Good Luck ☺