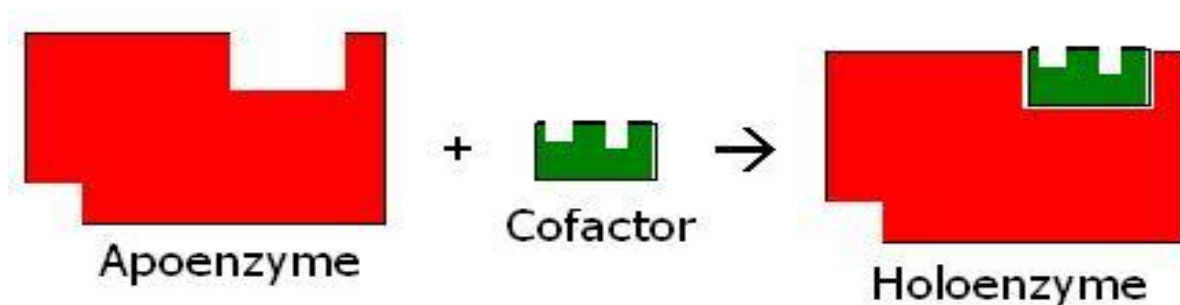


## DEFINITION OF VARIOUS TERMS USED IN ENZYMES WITH EXAMPLES:

1. **HOLOENZYME:-** An apoenzyme together with its cofactor. A holoenzyme is complete and catalytically active. Most cofactors are not covalently bound but instead are tightly bound. However, organic prosthetic groups such as an iron ion or a vitamin can be covalently bound. Examples of holoenzymes include DNA polymerase and RNA polymerase which contain multiple protein subunits. The complete complexes contain all the subunits necessary for activity.

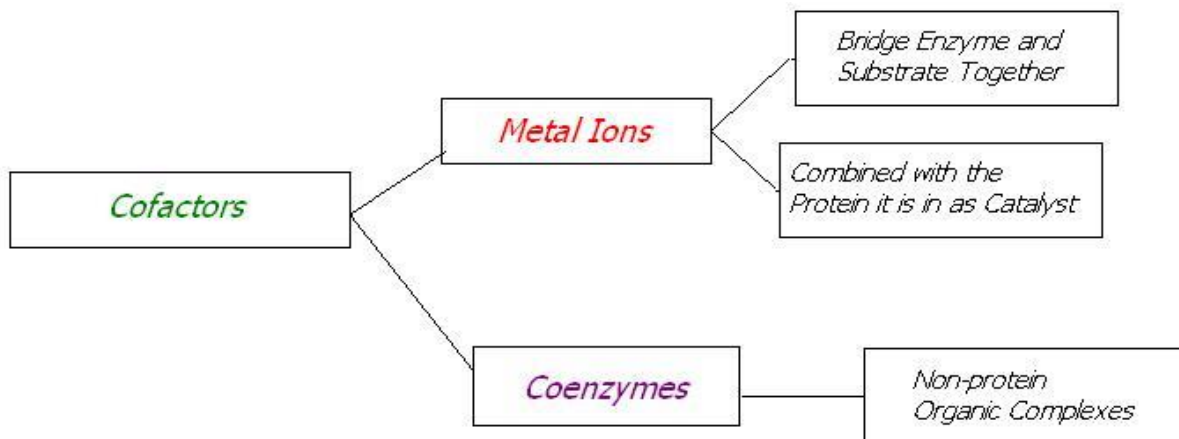


- Examples of Holoenzymes:
- DNA polymerase is a holoenzyme that catalyzes the polymerization of deoxyribonucleotides into a DNA strand. DNA polymerase is an active participant in DNA replication. It reads the intact DNA strand as a template and uses it to synthesize the new strand. The newly polymerized DNA strand is complementary to the template strand and identical to template's original partner strand. DNA polymerase uses a magnesium ion for catalytic activity.
- RNA polymerase is also a holoenzyme that catalyzes RNA. RNA polymerase is needed for constructing RNA chains from DNA

genes as templates, a process known as transcription. It polymerizes ribonucleotides at the 3' end of an RNA transcript.

2. **APOENZYME:-** An enzyme that requires a cofactor but does not have one bound. An apoenzyme is an inactive enzyme, activation of the enzyme occurs upon binding of an organic or inorganic cofactor.
3. **COFACTOR:-** Many enzymes require an additional small molecule, known as a cofactor to aid with catalytic activity. A cofactor is a non-protein molecule that carries out chemical reactions that cannot be performed by the standard 20 amino acids. Cofactors can be either inorganic molecules (metals) or small organic molecules (coenzymes).

Cofactors, mostly metal ions or coenzyme, are inorganic and organic chemicals that function in reactions of enzymes. Coenzymes are organic molecules that are nonproteins and mostly derivatives of vitamins soluble in water by phosphorylation; they bind apoenzyme protein molecule to produce active holoenzyme.



**4.COENZYMES** : The non-protein, organic, low molecular weight and dialyzable substance associated with enzyme function is known coenzyme.

- Coenzymes are nonprotein organic molecules that bind loosely to an enzyme. Many (not all) are vitamins or are derived from vitamins. Many coenzymes contain adenosine monophosphate (AMP). Coenzymes may be described as either cosubstrates or prosthetic groups.

Function: coenzymes are frequently required in oxido-reduction, group transfer and isomerization reaction and reaction resulting in the formation of covalent bond.

Coenzymes frequently contain vitamin B complex on part of their structures

For ex- thiamine (vit B1), riboflavin(vit B2), Lipoic acid, folic acid and vit B12 are part of several imp coenzymes involved in several biochemical reactions.

Classification:

1. Coenzymes involved in hydrogen transfer from one substrate to another.

- $\text{NAD}^+$  and  $\text{NADP}^+$
- FMN and FAD
- Lipoic acid
- Coenzyme Q

2. Coenzyme involved in transfer of group or atom other than hydrogen.

- ATP and related compounds
- Sugar phosphates
- Coenzyme A

- Folate coenzymes
- Vit B12
- Lipoic acid

5. **COSUBSTRATES** are coenzymes that bind tightly to a protein, yet will be released and bind again at some point.

6. **PROSTHETIC GROUPS** are enzyme partner molecules that bind tightly or covalently to the enzyme (remember, coenzymes bind loosely). While cosubstrates bind temporarily, prosthetic groups permanently bond with a protein. Prosthetic groups help proteins bind other molecules, act as structural elements, and act as charge carriers. An example of a prosthetic group is heme in hemoglobin, myoglobin, and cytochrome. The iron (Fe) found at the center of the heme prosthetic group allows it to bind and release oxygen in the lung and tissues, respectively. Vitamins are also examples of prosthetic groups.

- **Apoenzyme** is the name given to an inactive enzyme that lacks its coenzymes or cofactors.
- **Holoenzyme** is the term used to describe an enzyme that is complete with its coenzymes and cofactors.
- **Holoprotein** is the word used for a protein with a prosthetic group or cofactor.

7. **ACTIVE SITE:** The specific region of an enzyme where a substrate binds and catalysis takes place or where chemical reaction occurs.

Active site is made up of very specific sequence of amino acids determined by genetic codes.

The active site (or active centre) of an enzyme represents as the small region at which the substrate(s) binds and participates in the catalysis.

Salient features of active site

- 1 . The existence of active site is due to the tertiary structure of protein resulting in three dimensional native conformation.
2. The active site is made up of amino acids (known as catalytic residues) which are far from each other in the linear sequence of amino acids (primary structure of protein). For instance, the enzyme lysozyme has 129 amino acids. The active site is formed by the contribution of amino acid residues numbered 35, 52, 62, 63 and 101.
3. Active sites are regarded as clefts or crevices or pockets occupying a small region in a big enzyme molecule.
4. The active site is not rigid in structure and shape. It is rather flexible to promote the specific substrate binding.
5. Generally, the active site possesses a substrate binding site and a catalytic site. The latter is for the catalysis of the specific reaction.
6. The coenzymes or cofactors on which some enzymes depend are present as a part of the catalytic site.
7. The substrate(s) binds at the active site by weak non covalent bonds.
8. Enzymes are specific in their function due to the existence of active sites.
9. The commonly found amino acids at the active sites are serine, aspartate, histidine, cysteine, lysine, arginine, glutamate, tyrosine etc. Among these amino acids, serine is the most frequently found.

10. The substrate (S) binds the enzyme (E) at the active site to form enzyme-substrate complex (ES). The product (P) is released after the catalysis and the enzyme is available for reuse.

8. UNITS OF ENZYME ACTIVITY: enzyme unit, or international unit for enzyme (symbol U, sometimes also IU) is a unit of enzyme's catalytic activity.

1 U ( $\mu\text{mol}/\text{min}$ ) is defined as the amount of the enzyme that catalyzes the conversion of one micromole of substrate per minute under the specified conditions of the assay method.

The specified conditions will usually be the optimum conditions, which including but not limited to temperature, pH, and substrate concentration, that yield the maximal substrate conversion rate for that particular enzyme. In some assay method, one usually takes a temperature of  $25^{\circ}\text{C}$ .

One katal is the enzyme activity that converts one mole of substrate per second under specified assay conditions.