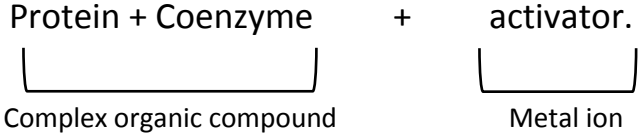
The enzymes:

- In the tissue of the plant, synthetic processes are continuously being carried on and at the same time sugar and other substances are continuously being broken down into simpler constitutes. The chemical reactions resulting in the synthesis of complex organic substances are known as plant **metabolism**. The synthesis of complex organic substances from raw materials is usually called **anabolism**, the breakdown of these substances to simpler, organic or in organic substances called **catabolism**. These reactions are generally absorbing energy or releasing energy.
- Enzymes are specific substances or organic catalysts highly active which their presence are necessary for energy consuming processes are occurring in the plant juice, this indicates that the plant juice contains enzymes that can catalyze the breakdown of the particular substances. Step by step to simpler substances and in each step amount of energy is released which may easily be used to drive an organic or energy absorbing reactions.
- Enzyme co-enzyme – activator relationships:
  Certain of enzymes consist only of proteins, like certain of proteolytic enzymes but other certain enzymes require the presence of some other compound to be enzymatically active. These compounds are non-portentous, terms; holo enzymes and apo-enzymes have also been component respectively. The non-protein component may be firmly attached to the protein when it is called a prosthetic group or it may be readily removed by dialysis in which case it is called coenzyme. The coenzyme or prosthetic group is heat stable, being unaffected by boiling.
- Non protein component if is a metal ion called activator, the protein is in active in absence of the metal ion.

  e.g. Tyrosinase enzyme which attach the amino acid tyrosine is made up from protein portion + an atom of copper (Cu).
- Many enzymes consist of three components:



Properties of enzymes:

1. Solubility: enzymes are soluble in water, dilute glycerin, alcohol, saline and buffer solution.
2. Specificity: the enzymes are able to piece together or to split up only with certain molecule. e.g.; Invertase enzyme which will act only on sucrose and not even on any other disaccharide, or may be an enzyme show specificity for a particular chemical linkage.
3. Catalytic activity.
4. Sensitivity: for heat (destroyed at 60-100^{o}c) or exposing to light (particularly to ultra violet) and also for variations in pH, and presence of certain organic salts.

- **Factors affected on enzyme kinetic:**
  1. Enzyme concentration.
  2. Substrate concentration.
  3. Temperature effect.
  4. Hydrogen ion concentration (pH).
  5. Activators and inhibitors.
  6. Accumulation of reactions products.
  7. The water.
- **Classification of enzymes**:

This classification is generally based on the nature of the reactions; they catalyse rather one the nature of the enzyme itself. They classified into six classes:

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

Practice part:

Detection of certain respiratory enzymes in plant tissues:

1. Detection of oxidation enzymes (Dehydrogenase)

- **Materials**: thin layers of Potato tubers, test tubes boiling water bath, methyl blue solution 0.025%.
- **Procedures**:
  1. Put some number of potato layers in two test tubes.
  2. Put one of them in boiling water bath for (10 mints.) then cool it.
  3. Full the two test tubes with methyl blue without bulb and close them.

4. After (24 hrs.) in library temperature, we notice that which one not putted in water bath, colored with returned to colorless, why?.

2. Detection of catalase enzyme:

Enzymes are found in plant tissues and microorganisms that can analysis the hydrogen peroxide portion (harmful portion) inside the cells.

$$2H_2O_2 \longrightarrow 2H_2O + O_2$$

- Materials: Potato tubers this layers, H_2O_2.
- Procedures:

1. Put the tuber layers in the pitrydish.
2. Adding the liquid of H_2O_2 (hydrogen peroxide).
3. Noticed that oxygen bulbs get out from the putted before that in boiling water for (10 mints.), what you see?.

3.Extraction and detection of Invertase enzyme:

The enzyme found in the plant parts and also in the microorganisms like Yeast.

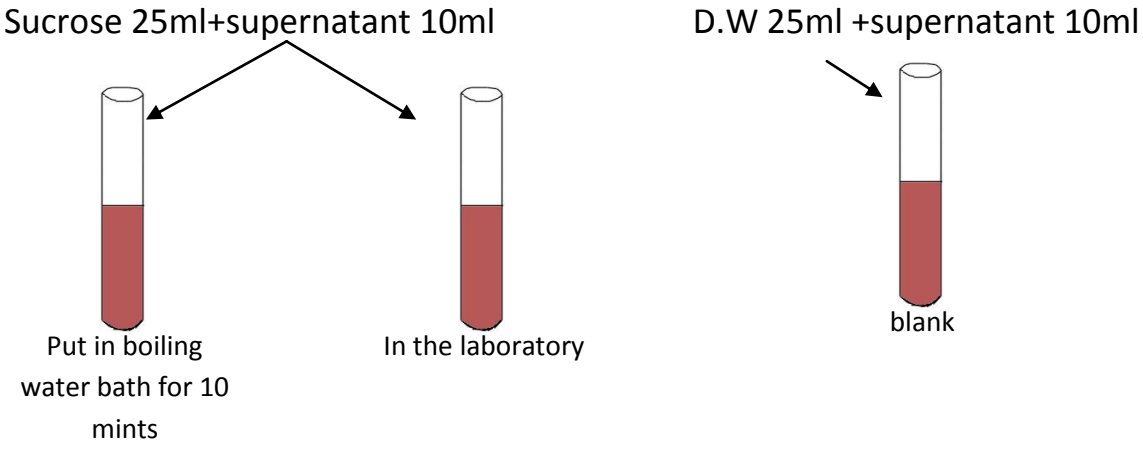
Sucrose glucose + fructose

Materials:

Dry yeast, sucrose (2%), boiling water bath, antifungal, test tubes, beakers, Benedict's reagent and cylinder.

Procedures:

1. (2gm) of yeast squish to powder.
2. Adding (40 ml) distal water inside a beaker.
3. Leave it for (20mints).
4. Centrifuges process doing for (5 mints/5000 circle) then collect or remove the supernatant.
5. Put (10ml) from the supernatant in a test tube (3 test tubes).
6. Adding (25ml) of sucrose solution.



7. Take (2ml) from each one of test tubes and add Benedict's solution to them. Notice any return in color, in each one the indicator return the color, why?

- Benedict's solution or reagent/ to detect for mono or disaccharides,
  blue color yellow or colorless.

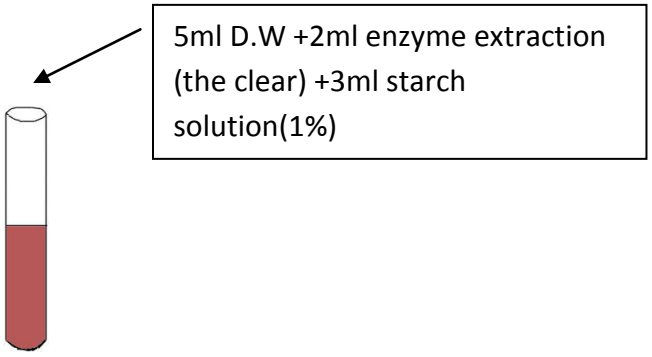
4. Extraction and action of the enzyme amylase:

Starch glucose.

Materials:

Cereal grain (wheat or barley) 20gm, starch solution (1%), Benedict's reagent, boiling water bath and test tubes.

Procedure:

1. Squish the grains to get out the enzyme with amount of distal water.
2. The mixture filtrated with clean shahs.
3. Centrifuge process (5 mints/5000 circle) for supernatant, then kept the clear in refrigerator.
4. 

The test tube put in water bath with laboratory temp. to increase the reaction velocity. Then this detection conducted after (15,30 mint).

5. The detection --- 1 ml from test tube content + 2 ml Benedict's solution.

6.The result ------ color return from **blue to yellow** indicated that the glucose found, that's resulted from starch analysis with enzyme of amylase.