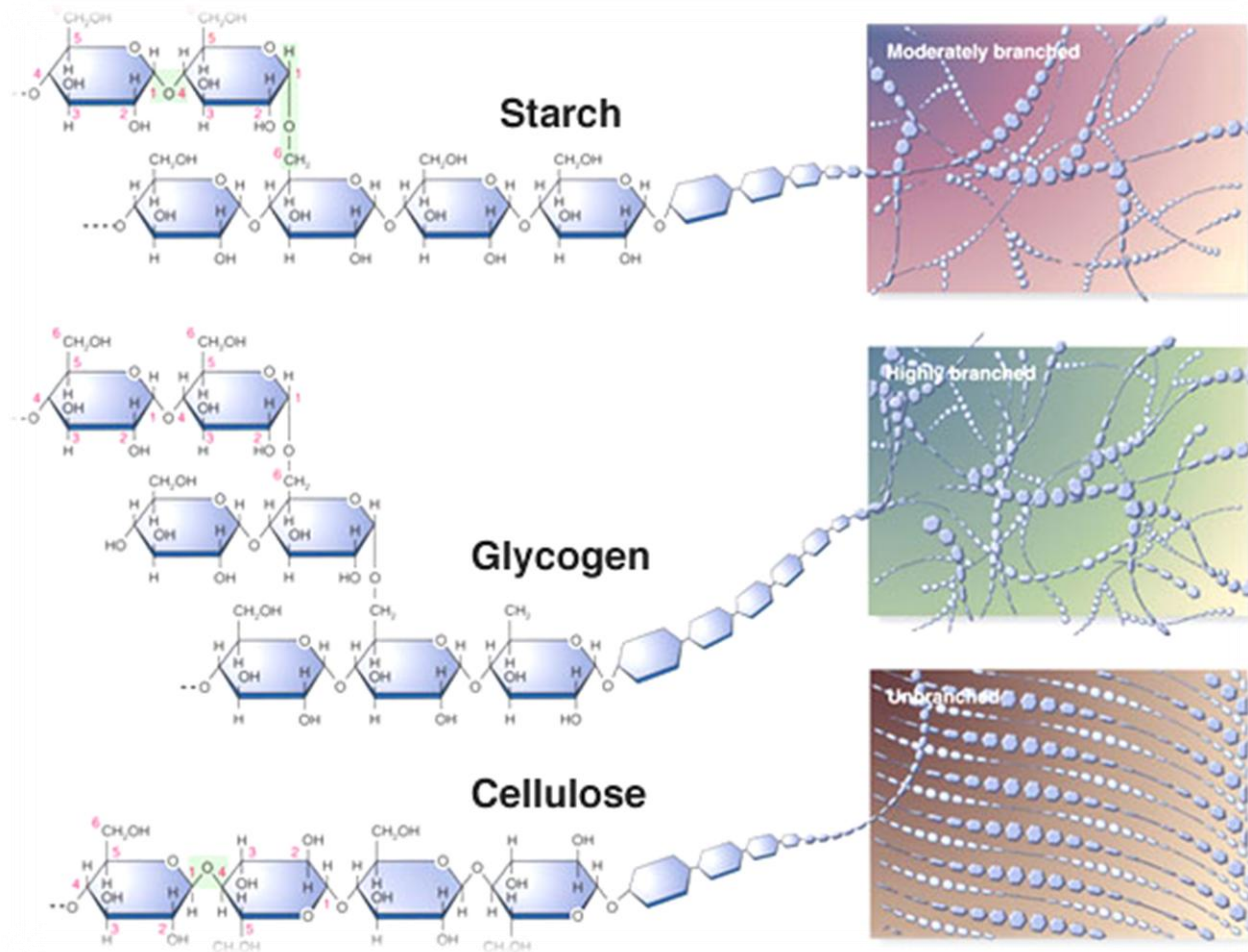


Qualitative analysis of Carbohydrates II

By : Amal Alamri

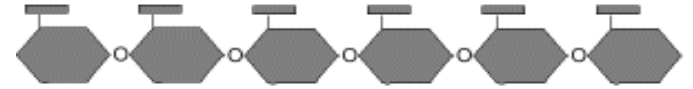


Polysaccharides

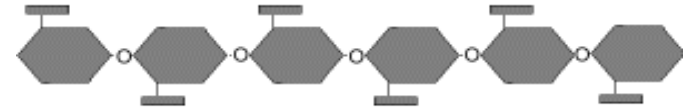
Polysaccharides can either be **homopolymeric** (same repeating monosaccharide unit) or **heteropolymeric** (mixture of monosaccharides).

Disaccharides can be broken into two monosaccharide units by hydrolysis and lost one molecule of water, examples of disaccharides Sucrose, Lactose and Maltose.

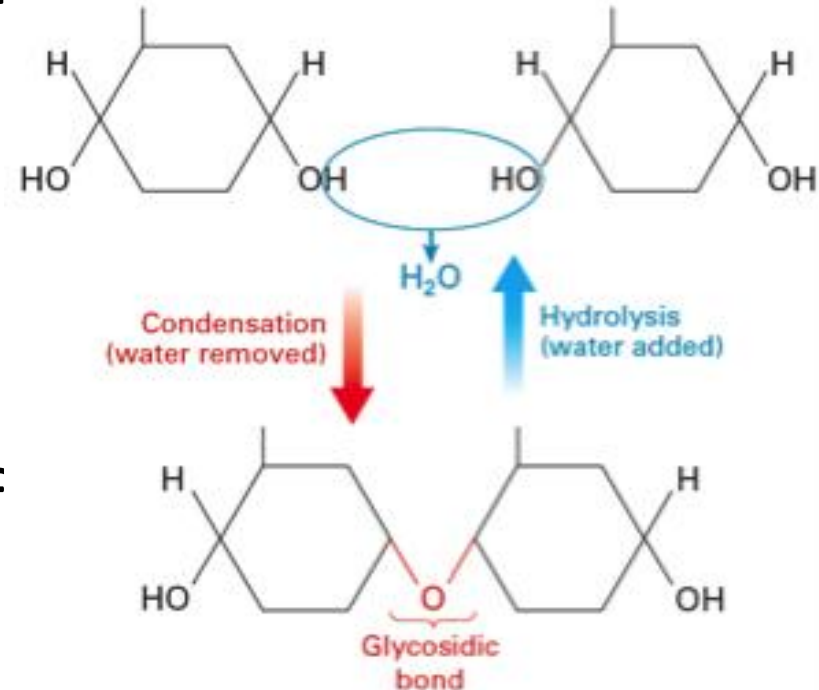
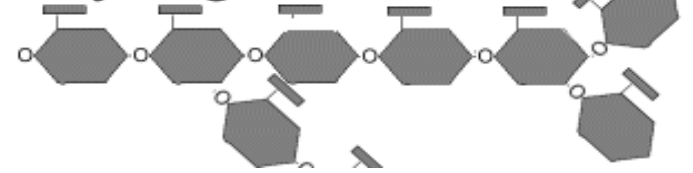
Starch



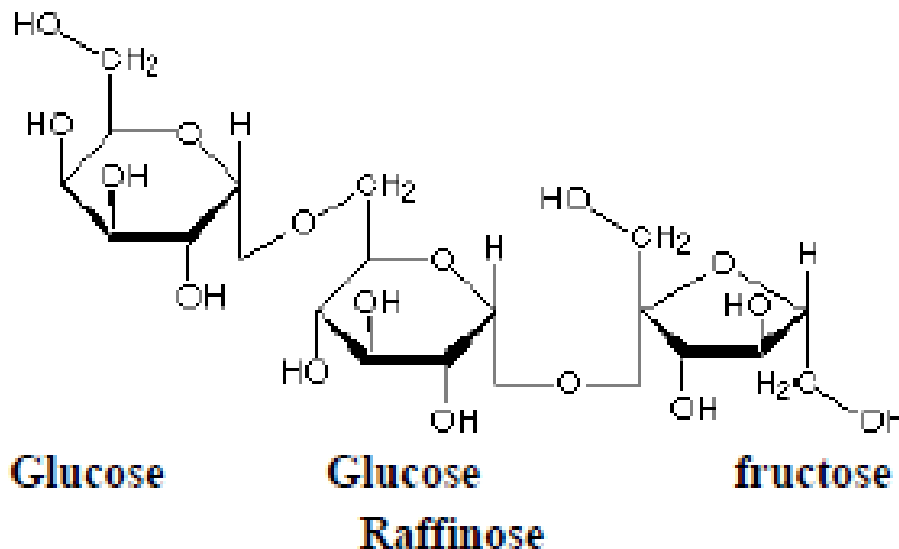
Cellulose



Glycogen



Oligosaccharides can be broken into 3-6 monosaccharide units by **hydrolysis** and **lost** one or more molecules of water.



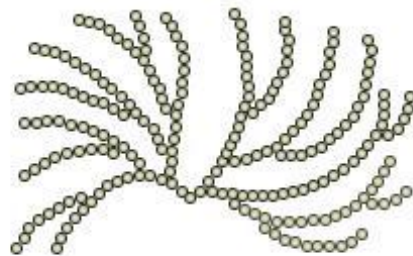
Polysaccharides

- Plants and animals store glucose in the form of very large polysaccharide glucose **homopolymers** that contain **both α 1-4 and α 1-6 glycosidic bonds**.
- The glucose homopolymer produced in plants is called starch, while the glucose homopolymer produced in animal cells is called glycogen.**

Plants synthesize **two forms of starch**, **amylose**, a **linear polysaccharide** containing about ~ 100 glucose units **linked by $\alpha(1-4)$ glycosidic bonds**, and **amylopectin**, a **branched polysaccharide** containing $\sim 100,000$ glucose units **connected by $\alpha 1-4$ and $\alpha 1-6$ glycosidic bonds**.

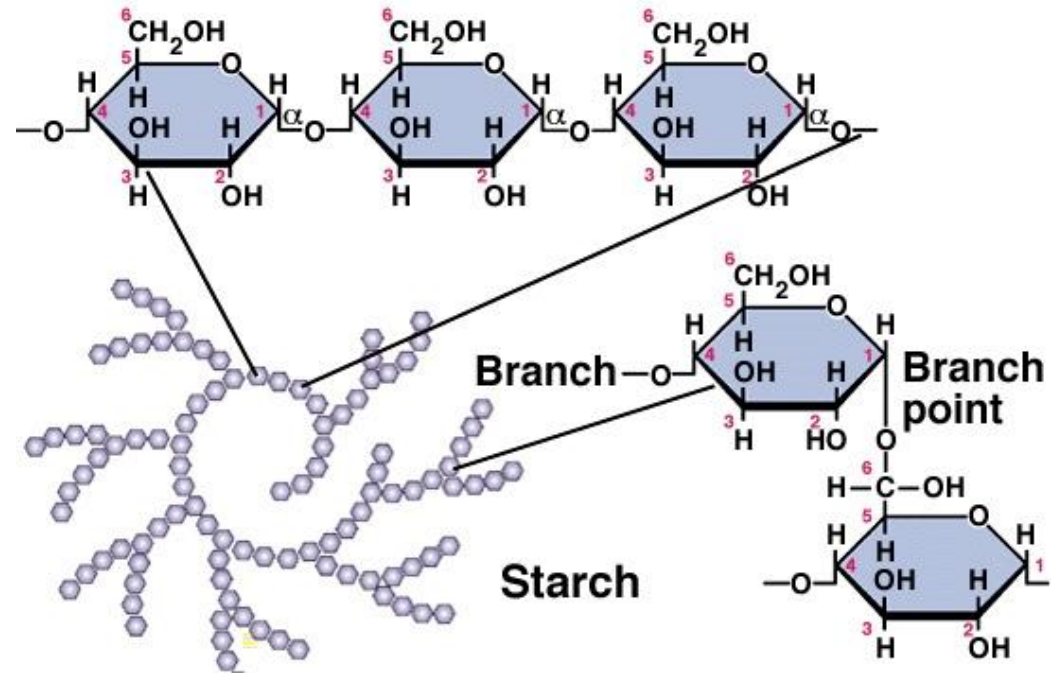


Amylose



Amylopectin

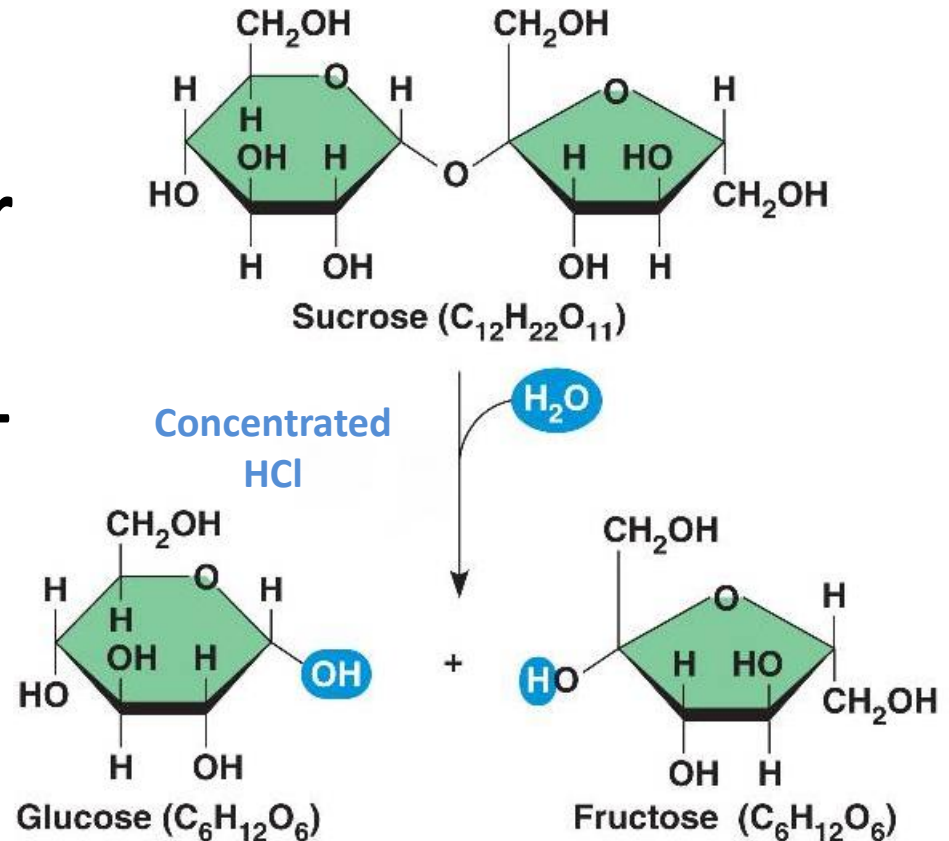
Polysaccharides



Experiment 1 : Sucrose Hydrolysis Test

Objective:

This test is used to convert sucrose (non-reducing disaccharide) to glucose + fructose (reducing monosaccharides).

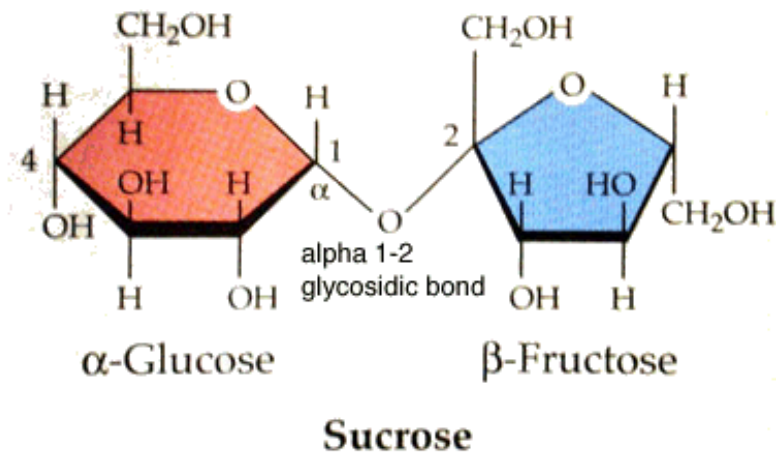


Principle:

-Sucrose is the only non-reducing disaccharide so it does not reduce the Cu^{++} solution (Benedict's and Fehling's test) because the glycosidic bond is formed between the two hemiacetal bonds.

-So there is no free aldehydic or ketonic group to give positive reducing properties.

This bond can be hydrolysed by strong acid ; concentrated HCl and the individual components of sucrose (glucose + fructose) are then able **to give positive reducing test.**



Materials:

Sucrose

Concentrated hydrochloric acid (HCl)

Benedict's reagent

Seliwanoff's reagent

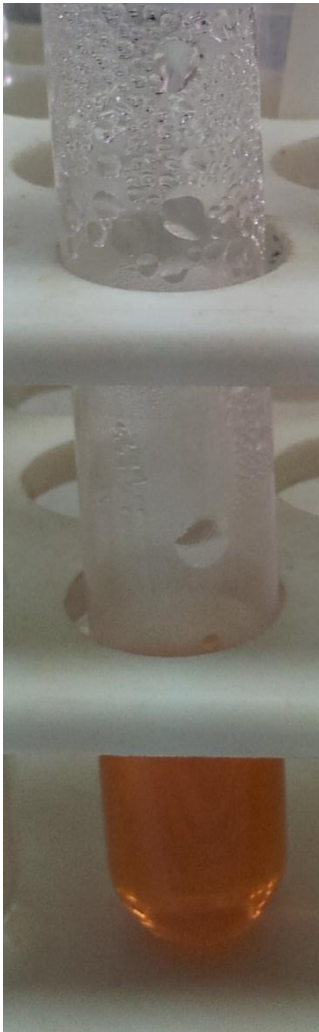
Method:

- 1- Set up two tubes add to each one 4ml of a sucrose solution ,Label the tube : (**Sucrose with HCl, Sucrose without HCl**)
- 2- **To only one tube** add four drops of concentrated hydrochloric acid (HCl)
- 3-**Heat both in boiling water bath for 15 minutes.**
- 4- After 15 minutes of heating **add 4 drops of concentrated NaOH** to each tube (?)
- 5-**From the tube containing HCl take 2ml in two tubes** to do Benedict's test and Seliwanoff's test , label the tube (Benedict +HCl) and (Seliwanoff'+HCl)
Add 2 ml of Benedict's reagent and 2.5 ml of Seliwanoff's reagent **WHAT do expect?**
- 6-**From the tube which contains only sucrose take 2 ml to do Benedict's test only** (add 2 ml of Benedict's reagent) **WHAT do expect?**

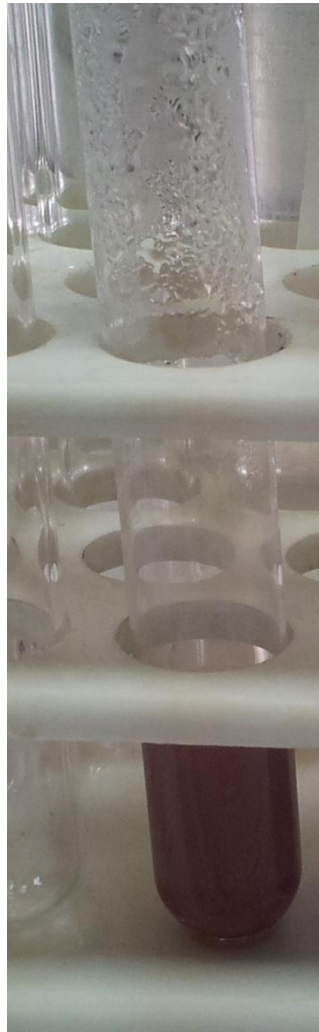
Result :

Sucrose with HCL		Sucrose without HCL
Benedict's test	Seliwanoff's test	Benedict's test

Write your observation and discuses each result.



Sucrose+ HCl
Seliwanoff's test (+)



Sucrose+ HCl
Benedict's test(+)



Sucrose only
Benedict's test(-)

Experiment 2: The Iodine/Potassium Iodide Test

Objective:

This test used to distinguish between polysaccharides and mono or oligo saccharides.



Principle:

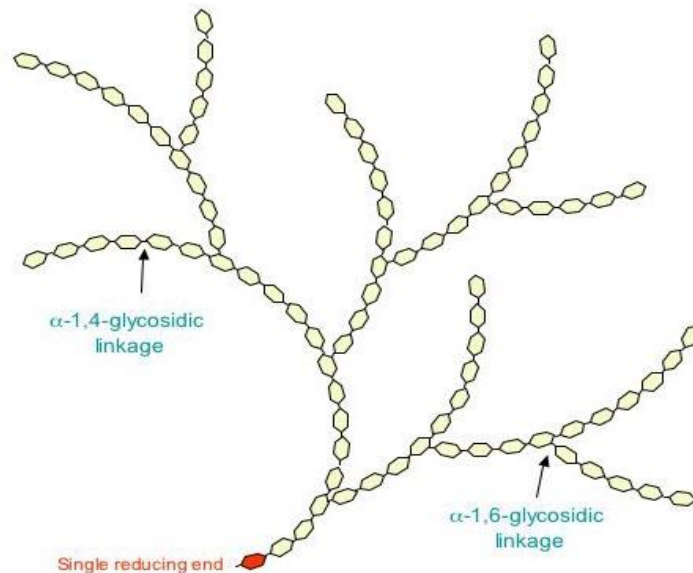
Starch forms deeply blue color complex with iodine.

Starch contains : α - amylose, a helical saccharide polymer and amylopectin.

Iodine forms a large complex with α -amylose helix.

- Simple oligosaccharides and mono saccharides do not form this complex.

-Note that other polysaccharides like glycogen may give other colors (red).

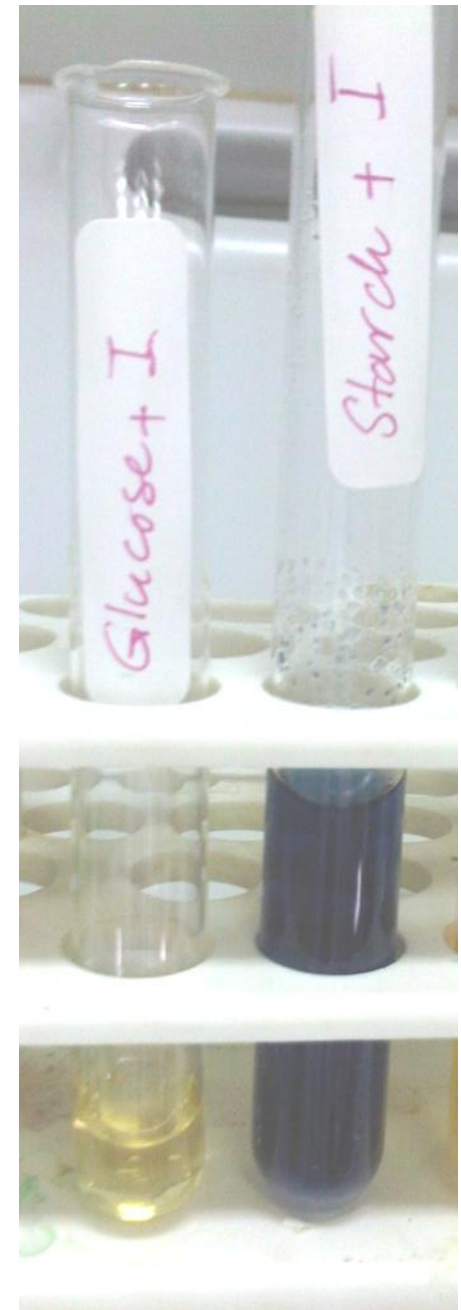


Structure of amylopectin, a branched starch



About the test...

The blue colour can be detected visually with concentrations of iodine as low as 0.00002M at 20°C . **However the intensity of the colour decreases with increasing temperature.** Also the test cannot be done at very low pHs **due to the hydrolysis of the starch under these conditions.**



Materials:

Iodine/potassium iodide solution

Starch, glucose

Method:

- Two ml of a sample solution is placed in a test tube.
- Add 2drops of iodine solution and one ml of water. Shake it well
- A positive test is indicated by the formation of a **blue-black complex**.
- **Take a half of the tube of starch and heat it in boiling water bath for 10 min compare between the tow tubes and write your observation.**

Result :

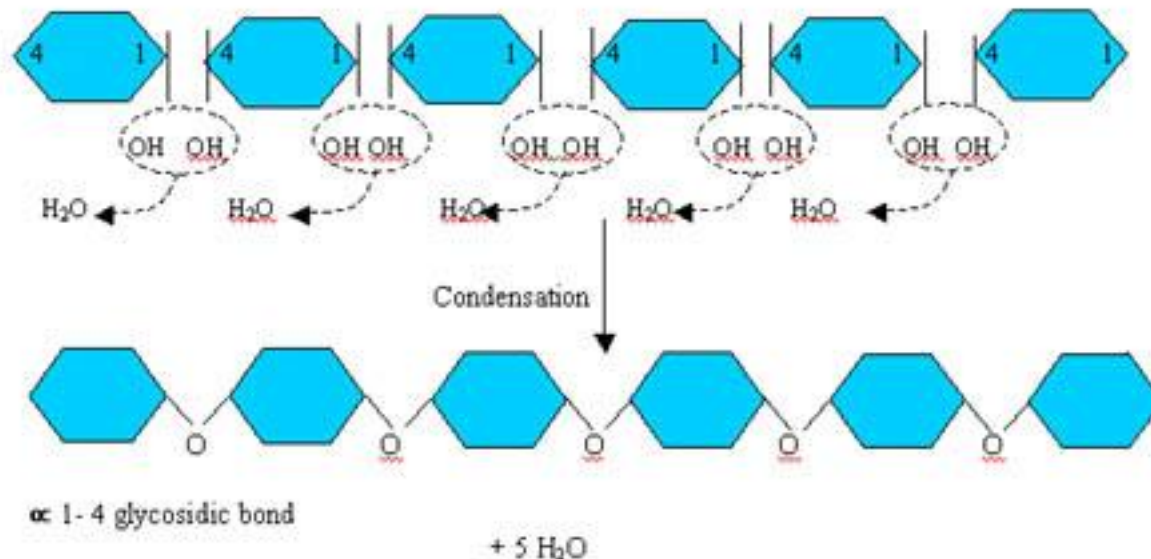
Tube	Observation	Comments
(Starch + Iodine) without heating		
(Starch + Iodine) after heating		
(Glucose+ Iodine)		

Write your observation and discuses each result.

Experiment 3: Hydrolysis of Starch:

Objective :

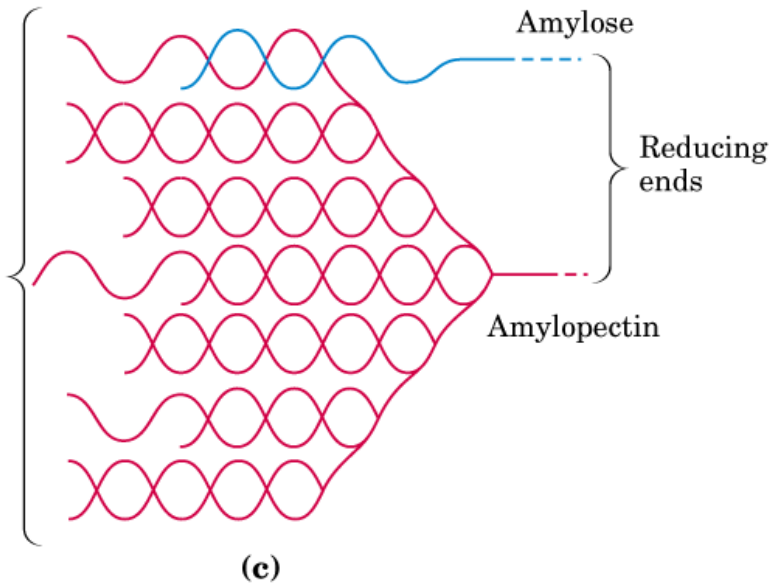
This experiment illustrates the conversion of starch (non-reducing sugar) to a reducing sugar by the action of hydrochloric acid at boiling point. The longer the starch is exposed to the acid the further hydrolysis proceeds



Principle:

-Although starch has free hemiacetal in the terminal glucose residue, it has no reducing properties, because the percentage between the free residues is very low in comparison to the whole molecule.

-Heating starch solution in acid medium hydrolyses the glycosidic bonds giving many free glucose residues. These glucose molecules give reducing properties to the hydrolysis product.



Materials:

Starch

Benedict's reagent

iodide solution

Water bath

Method:

1-Two ml of starch in large tube

2- Add three drops of Hydrochloric acid, heated in boiling water bath for 10 mints. then cold solution

3-Add the amount of sodium hydroxide to become the base

4-Divided in two tube (a,b)

5- In tube (a) add 1 ml of iodine solution and note the result. **WHAT do expect?**

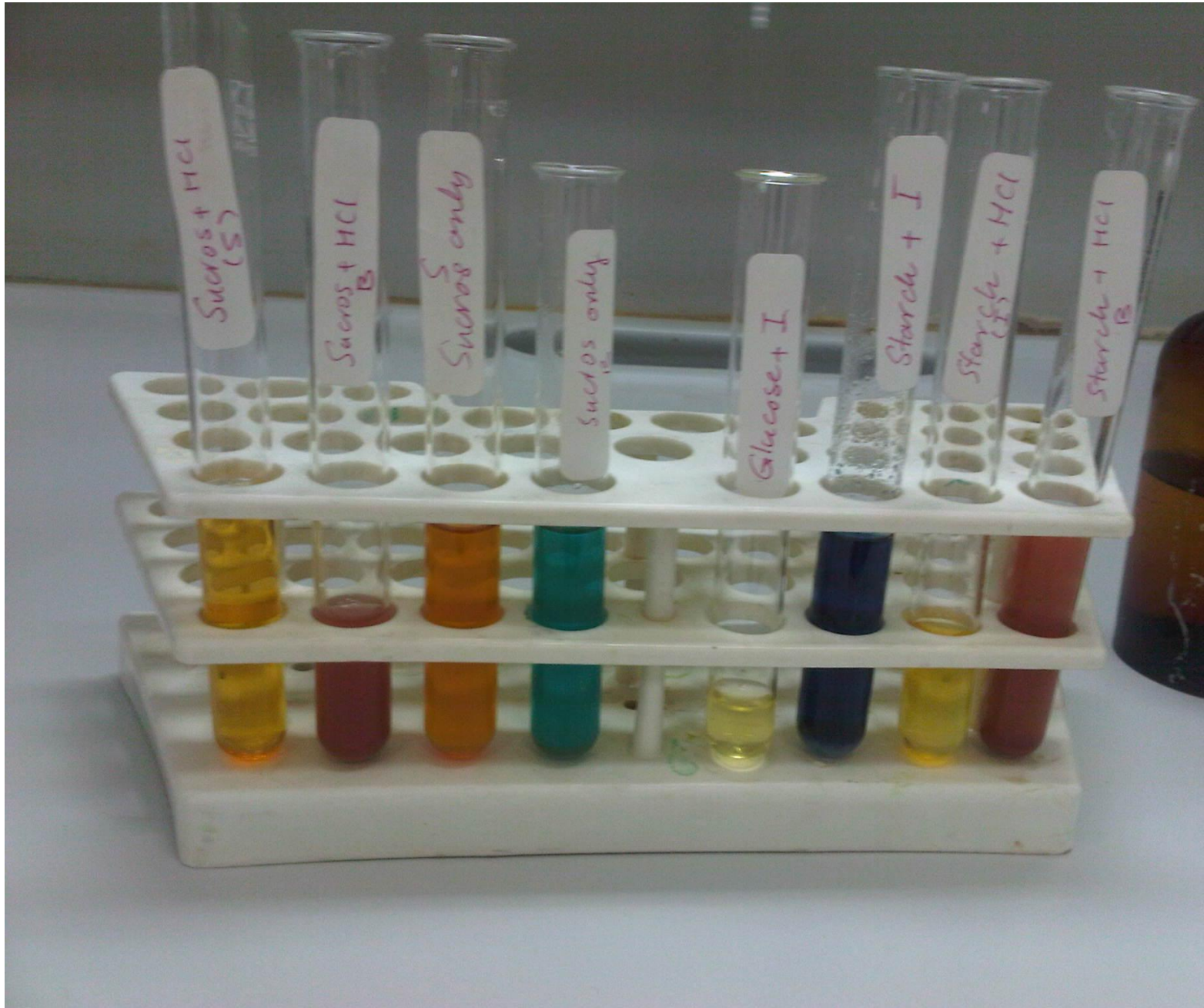
6- In tube (b) add 1 ml of Benedict reagent, mix and heated for 3 mint and record result. **WHAT do expect?**

Result:

Starch with HCL	
Benedict's test	Iodine test

Write your observation and discuss each result.





THANK YOU 😊