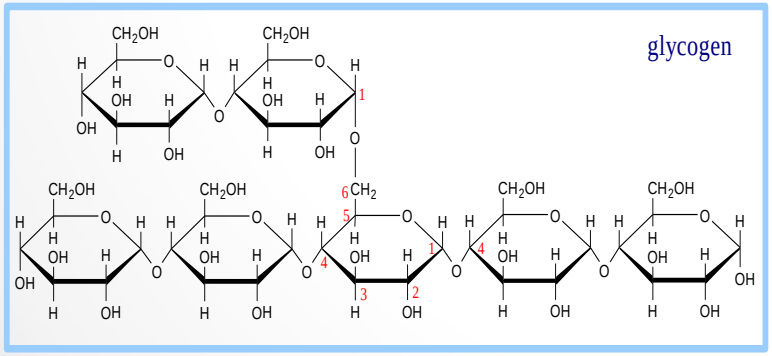
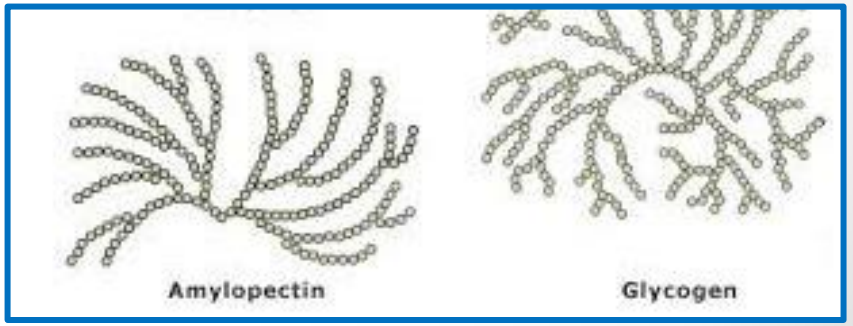
Qualitative analysis of carbohydrates II





Complex carbohydrate

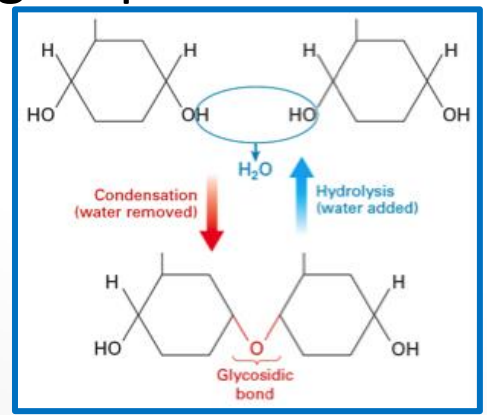
Complex sugars consist of more than one unit of monosachride, it could be:

-Disaccharides contain **two** monosaccharide units.

- Oligosaccharides contain **3-9** monosaccharide units.

- Polysaccharides can contain more than 9 monosaccharide units.

- Complex carbohydrates can be broken down into smaller sugar units through a process known as **hydrolysis**.



Complex carbohydrate

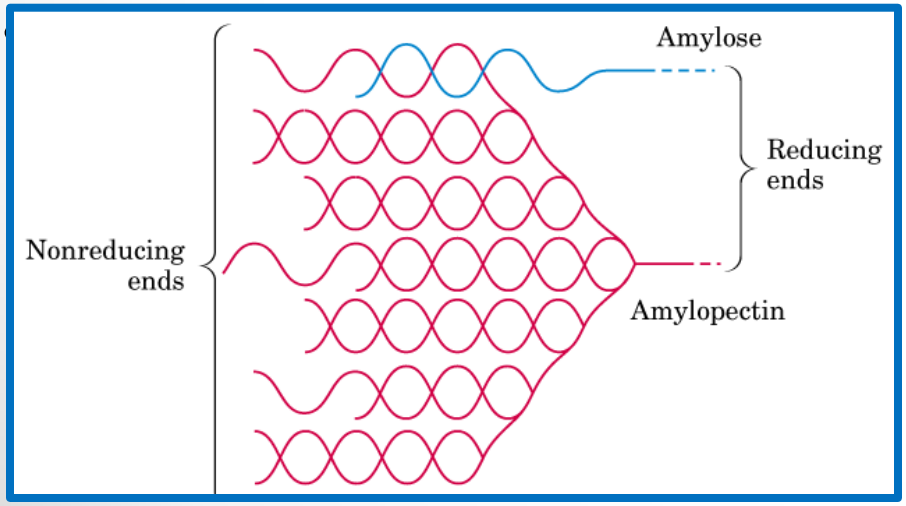
- Polysaccharides can either be homopolymeric (same repeating monosaccharide unit) or heteropolymeric (mixture of monosaccharaides).

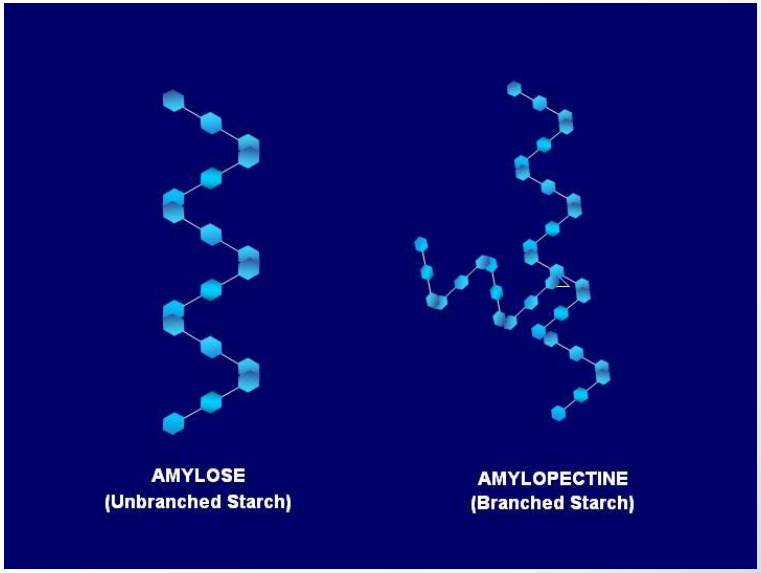
- Plants and animals store glucose in the form of very large polysaccharide glucose **homopolymers** .

- The glucose homopolymer produced in plants to store glucose is called starch, while the glucose homopolymer produced in animal cells is called glycogen.

Polysaccaride

- **Starch consists of** Two forms: amylose and amylopectin ..
- **Amylose is a** Linear helical polymer of glucose linked by α-1,4 glycosidic bonds (100 units)
- **Amylopectin** Branched polymer containing glucose linked by α-1,4 glycosidic bonds. Branch points has α-1,6glycosidic bonds, (100,000 units)

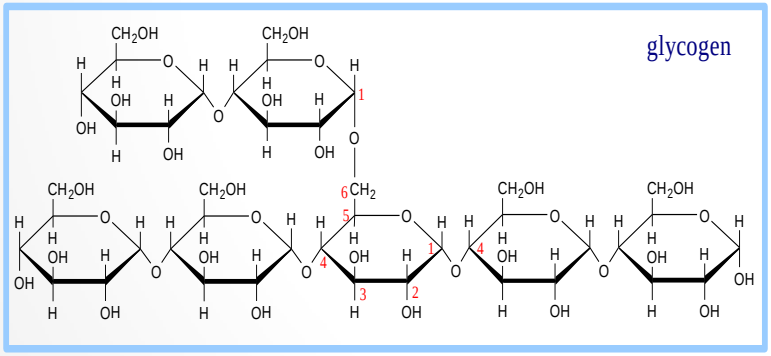


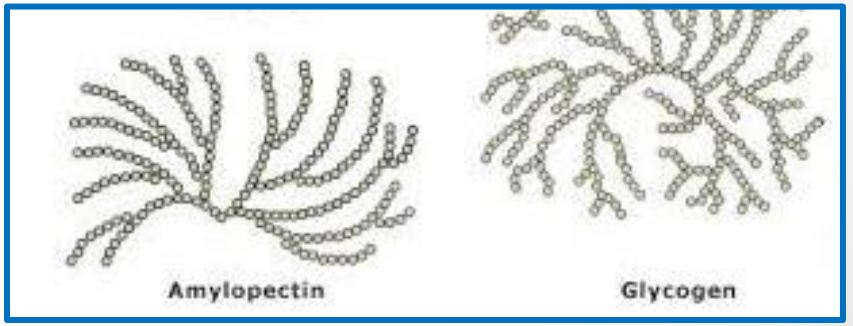


Glycogen

Glycogen, is a branched polysacharide of D-glucose which contains both α(1→4) and α(1→6) is similar in structure to amylopectin.

But glycogen has **more α(1→6) branches**.



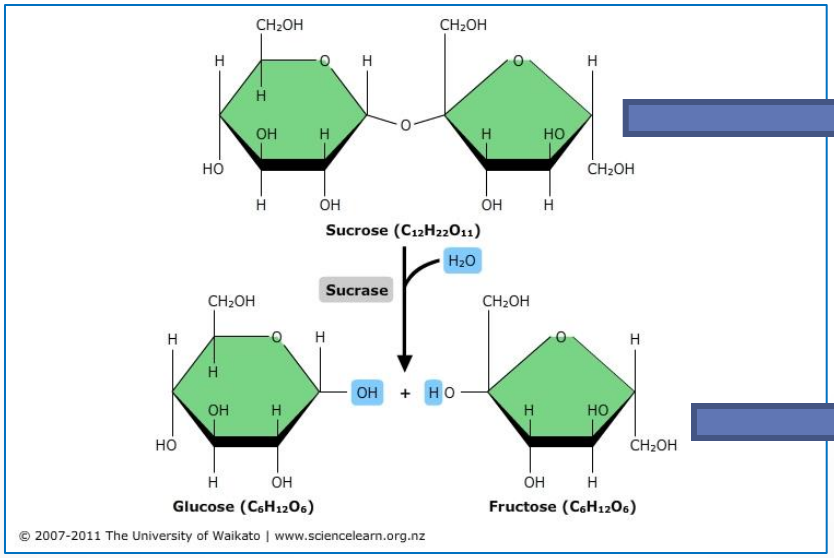


1-Sucrose hydrolysis Test

- This test is used to convert sucrose (non-reducing disaccharide) to glucose and fructose (reducing mono saccharides).
- **Objective**: To identify the products of hydrolysis of di- and polysaccharides.

Principle

- Sucrose is the only non-reducing disaccharide so it does not reduce the Cu++ solution (Bendict'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 and the individual components of sucrose (**glucose + fructose**) are then able to give positive reducing test.



No result with bendict

Positive result with bendect

Method:

1- Set up tow tubes add to each one 4ml of a sucrose solution ,Lable 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 tow tube to do Benedict's test and Seliwanoff's test , lable 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 contian only sucrose take 2 ml to do Benedict's test only (add 2 ml of Benedict's reagent) **WHAT do expect?**

Result

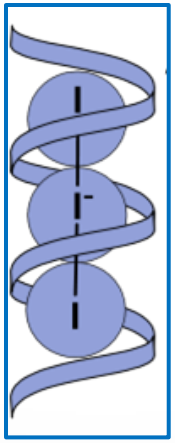
Sucrose with HCL		Sucrose without HCL
Benedict's test	Seliwanoff's test	Benedict's test

2-The Iodine/Potassium Iodide Test

- This test used to distinguish between polysaccharides and mono or oligo saccharides.
- **Objective**: to detect the presence of starch in a sample

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. This complex absorbs light and reflects the blue light only. Simple oligosaccharides and mono saccharides do not form this complex.
- **Note that other polysaccharides like glycogen may give other colors (red).**



- Amylopectin (and glycogen) are unable to assume a stable helical conformation because of the branching.
- Amylopectin complexes with iodine to a much lesser extent than amylose, therefore, the **amylopectin-iodin** complex has a red voilet color that is much less intense than blue of the **amylose-iodine** complex.

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.**

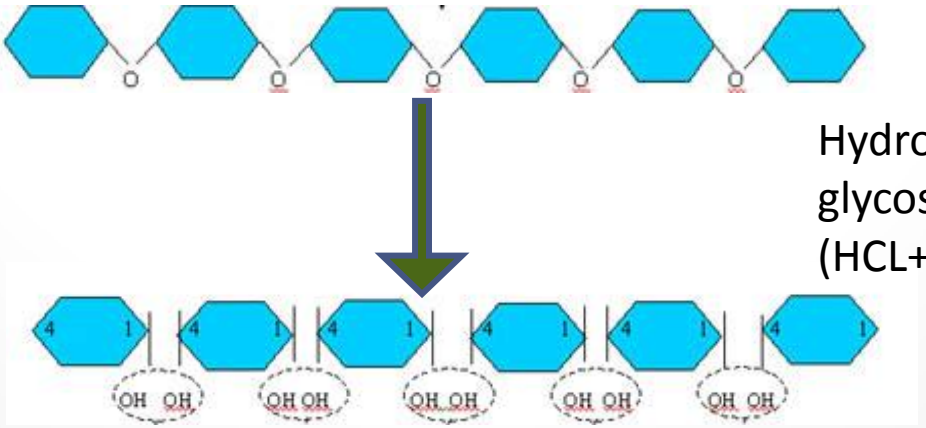
Result

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

3-Hydrolysis of Starch

- 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.

- Objective: to establish the effect of concentrated HCL on a glycosidic bond in starch.



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.

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