Dr. Dalys S. Obaida/PhD Clinical Biochemistry
Hanadi T. Muslim /MSc Biochemistry

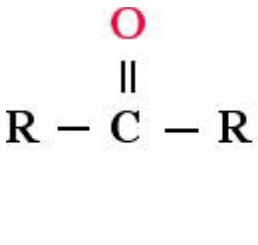
Carbohydrates tests

Introduction:

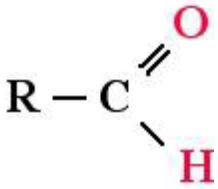
The word carbohydrate is formed from the words carbon and hydrogen.Carbohydrates are combinations of the chemical elements carbon and hydrogenplus oxygen. In the natural world, carbohydrates are the most common chemicalcompounds used for food. Carbohydrates are the most abundant and diverseclass of organic compounds occurring in nature. It played a key role in theestablishment and evolution of life on earth by creating a direct link between thesun and chemical energy. Carbohydrates are the key source of energy used byliving things.

•Also serve as extracellular structural elements as in cell wall ofbacteria and plant. Carbohydrates are defined as the polyhydroxy aldehydes orpolyhydroxy ketones.

•Most , but not all carbohydrate have a formula $(CH_2O)_n$ (hence the name
hydrate of carbon)
•In human body, the D-glucose is used.
•Simple sugars end with –ose.



Ketone Group



Aldehyde Group

Several classifications of carbohydrates have proven useful, and are outlined in the following table.

Complexity	**Simple Carbohydrates** monosaccharides			**Complex Carbohydrates** disaccharides, oligosaccharides & polysaccharides	
Size	**Tetrose** C_4 sugars	**Pentose** C_5 sugars	**Hexose** C_6 sugars	**Heptose** C_7 sugars	etc.
C=O Function	**Aldose** sugars having an aldehyde function or an acetal equivalent. **Ketose** sugars having a ketone function or an acetal equivalent.				
Reactivity	**Reducing** sugars oxidized by Tollens' reagent (or Benedict's or Fehling's reagents). **Non-reducing** sugars not oxidized by Tollens' or other reagents.				

Classification:

1-Simple sugar (one unit) :

Monosaccharides contain one monosaccharides unit

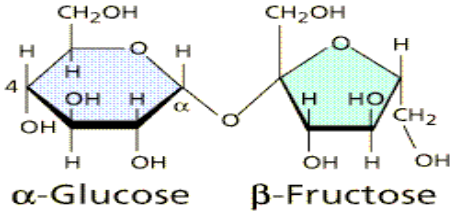
2-Complex sugar (more than one) :

•Disaccharides contain two monosaccharide units.

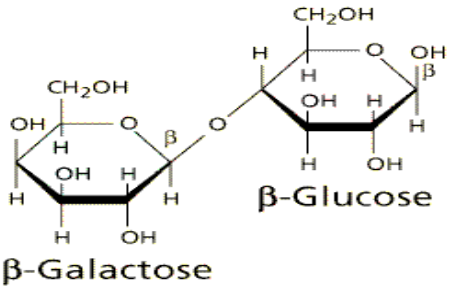
•Oligosaccharides contain 3-9 monosaccharide units.

•Polysaccharides can contain more than 9 monosaccharide units.

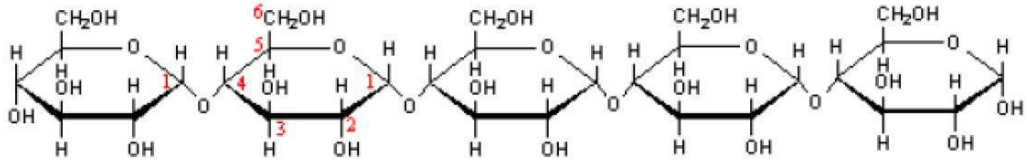
-Complex carbohydrates can be broken down into smaller sugar units through a process known as hydrolysis.



Sucrose



β-Lactose

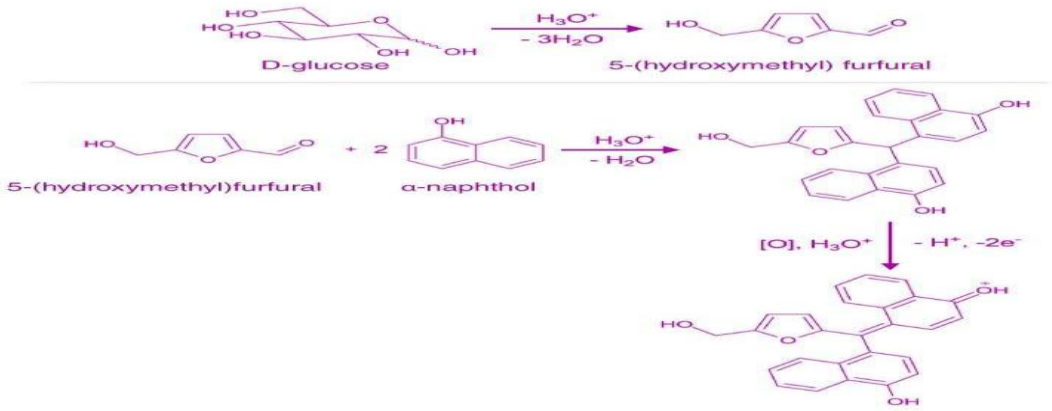


Amylose

Molisch Test: specific for carbohydrates.

Molisch test: Molisch's test is a chemical test which is used to check for the presence of carbohydrates in a given analyte. This test is named after Czech-Austrian botanist Hans Molisch, who is credited with its discovery. Molisch's test involves the addition of Molisch's reagent (a solution of ∝-naphthol in ethanol) to the analyte and the subsequent addition of a few drops of concentrated H2SO4 (sulphuric acid) to the mixture. The formation of a purple or a purplish-red ring at the point of contact between the H2SO4 and the analyte + Molisch's reagent mixture confirms the presence of carbohydrates in the analyte. This test is specific for all

carbohydrates Monosaccharide gives a rapid positive test, Disaccharides and polysaccharides react slower.



Objective: To identify the carbohydrate from other macromolecules, lipids and proteins. Principle: In Molisch's test, the carbohydrate (if present) undergoes dehydration upon the introduction of concentrated hydrochloric or sulphuric acid, resulting in the formation of an aldehyde.This aldehyde undergoes condensation along with two phenol-type molecules (such as ∝-naphthol, resorcinol, and thymol), resulting in theformation of a purple or reddish-purple colored complex.

•The test reagent(H2SO4) dehydrates pentose to form furfural and dehydrates hexoses to form 5- hydroxymethyl furfural

. •The furfural and 5-hydroxymethyl furfural further react with α-naphthol present in the test reagent to produce a purple ring.

Method:

1. 2-3 drops of Molisch's reagent (which α-napthol in 95% ethanol) must beaded to a small amount of the analyte in a test tube and mixed well.
2. few drops of concentrated sulphuric acid must be added dropwise along the walls of the test tube to facilitate the formation of a layer and avoid mixing. The development of a purple ring at the layer formed by the concentrated acid is a positive indicator for Molisch's test. If no purple or reddish-purple colour arises, the given analyte does not contain any carbohydrate.

The formation of a purple ring is a positive indicator for Molisch's Test