



وزارة التعليم العالي والبحث العلمي  
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كلية التقنيات الطبية  
قسم تقنيات المختبرات الطبية

## محاضرات الكيمياء الحياتية العملي

لطلبة تقنيات المختبرات الطبية  
المرحلة الاولى

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## Part one: Qualitative Analysis of Carbohydrates

### DEFINITION

Carbohydrates are defined as polyhydroxy alcohols with an aldehyde or ketone as the functional group.

### FUNCTIONS OF CARBOHYDRATES

1. Carbohydrates are the main source of energy in the body.
2. Storage form of energy (starch and glycogen)
3. Excess carbohydrate is converted to fat.
4. Glycoproteins and glycolipids are components of cell membranes and receptors.
5. They form structural basis of many organisms.

### CLASSIFICATION

Carbohydrates are classified according to the number of sugar molecules in them as monosaccharides, oligosaccharides and polysaccharides.

**Monosaccharides:** Monosaccharides are also called as simple sugars. They have only one potential sugar group. They consist of a single polyhydroxy aldehyde or ketone unit, and thus cannot be hydrolyzed into simpler form.

They may be subdivided into different groups as follows:

1. Depending upon the number of carbon atoms they possess, e.g.

<i>No. of carbon</i>	<i>Type of sugar</i>	<i>Aldoses</i>	<i>Ketoses</i>
1	Trioses	Glyceraldehyde	Dihydroxyacetone
2	Tetroses	Erythrose	Erythrulose
3	Pentoses	Ribose, xylose	Ribulose, xylulose
4	Hexoses	Glucose, galactose, mannose	Fructose
5	Heptoses	Glucoheptose	Sedoheptulose

2. Depending upon the functional groups:

- Aldehyde (CHO) - Aldoses                      **2**
- Ketone (C=O) - Ketoses.

**Disaccharides:** are classified as:

- *Reducing disaccharides:* In reducing disaccharides one of the functional groups is free. e.g. Maltose, Lactose

Type of oligosaccharide	No. of monosaccharide	Example	Type of monosaccharide present
Disaccharide	Two	Maltose Lactose Sucrose	Glucose + Glucose Glucose + Galactose Glucose + Fructose

*Non-reducing disaccharides:* Non-reducing disaccharides do not have free functional group. The potential functional groups are involved in glycosidic linkage. e.g. Sucrose.

Type of oligosaccharide	No. of monosaccharide	Example	Type of monosaccharide present
Trisaccharide	Three	Raffinose	Glucose + Galactose + Fructose
Tetrasaccharide	Four	Stachyose	2 molecules of Galactose + Glucose + Fructose
Pentasaccharide	Five	Verbascose	3 molecules of Galactose + Glucose + Fructose

**Oligosaccharides:** Oligosaccharides consist of a short chain of monosaccharide units (3 to 10 units), joined together by a characteristic bond called glycosidic bond. Oligosaccharides are subdivided into different groups based on the number of monosaccharide units present.

**Polysaccharides:** Polysaccharides are carbohydrates having more than ten monosaccharide units. They are also called glycans or complex carbohydrates.

They are classified into two types according to the type of monosaccharide units present.

1. Homopolysaccharides: Made up of repeated units of same type of monosaccharide units. e.g. Starch, glycogen, cellulose, inulin, dextrans, dextrans
2. Heteropolysaccharides: Made up of different types of monosaccharide units and their derivatives. e.g. Agar, gum, pectins, glycosaminoglycans such as hyaluronic acid, heparin sulfate, keratin sulfate, chondroitin sulfate.

### TESTS FOR CARBOHYDRATES

In order to understand and remember easily, the various tests of carbohydrates are explained in the beginning as general tests.

The various tests for carbohydrates are given below:

1. *Molisch test*: specific test for carbohydrates
2. *Iodine test*: specific test for polysaccharides
3. *Benedict's test*: specific test for reducing substances
4. *Barfoed's test*: specific test for monosaccharides
5. *Seliwanoff's test*: specific test for ketohexoses
6. *Osazone test*: to differentiate the reducing sugars on the basis of crystal formation.

### **Physical Properties**

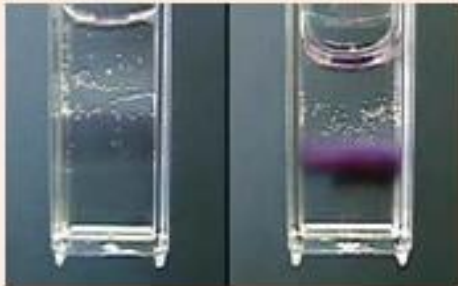
1. *Color*: Colorless except Starch which is pale white
2. *Clarity*: Clear except Starch which is cloudy
3. *Odor*: Odorless
4. *Reaction to litmus*: Neutral

### **Chemical Tests:**                      *Molisch Test:*

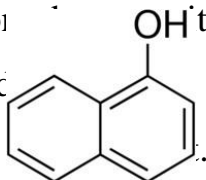
*Principle*: Carbohydrates, when treated with concentrated Sulfuric acid undergo dehydration to form furfural with pentose sugars / hydroxymethyl furfural for hexose sugars derivatives which on condensation with alphanaphthol form colored products.

*Molisch reagent*: A 5% solution of alpha naphthol in ethyl alcohol.

$\alpha$  – naphthol

Experiment	Observation	Inference
Take 2 ml of given solution in a clean dry test tube; add 1-2 drops of Molisch reagent. Mix. Incline the test tube slightly and overlay 2 ml of conc. sulphuric acid along the sides of the test tube so as to form two layers.	Violet ring at the junction of the two liquids is formed. 	Given solution is a carbohydrate.


### ***Points to Remember:***

- This is a general test for all carbohydrates.
- Molisch test is given by sugars with at least five carbon atoms. It involves furfural derivatives which are five carbon compounds.
- Impurities in the reagent tend to give a green ring, which is .
- A green ring even in absence of carbohydrates is due to excess of alpha naphthol.
- In case of oligo and polysaccharides, they are first hydrolyzed to monosaccharides by acid, which undergoes dehydration to form furfural or its derivatives.

### **Iodine Test:**

**Principle:** The test depends upon the property of adsorption possessed by the large polysaccharide molecules which adsorb the smaller iodine molecules on their surface to form the blue colored complex of ill-defined chemical nature. The property of adsorption decreases on heating, the complex dissociates and, therefore, the color disappears.

**Iodine reagent:** 0.5 ml of iodine diluted to 5 ml with distilled water.

<i>Experiment</i>	<i>Observation</i>	<i>Inference</i>
Take 2 ml of given solution in a test tube. Add 2-3 drops of Iodine solution.	Blue color is formed. 	Given solution is a polysaccharide.

**Points to Remember:**

- This is a specific test for polysaccharides.
- The amylose component of starch has a helical structure. When it is treated with iodine solution, Iodine is trapped inside the coil and the complex has an intense blue color. When the amylose solution is heated the helical conformation is disrupted and loses its capacity to bind iodine. On cooling the original conformation is regained and the capacity to bind iodine is also recovered.
- Sometimes the color may not reappear on cooling as small amounts of iodine added may vaporize away during heating.