

### MINISTRY OF HIGH EDUCATION AND SCIENTIFIC RESEARCH University of Ibn Khaldoun - Tiaret Institute of veterinarian sciences



# **Biochimical Polycopy**

	BOLISM		
Electro shuttle ac membra GLYCOLYSIS cose Pyruvate	ross 2 NADH	KREBS	ELECTRON TRANSPORT CHAIN AND OXIDATIVE PHOSPHORYLATIO
+ 2 ATP	- 0 to about 2 ATP	+ 2 ATP	+ about 34 ATP

Presented by : Bourabah Akila

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#### Introduction

Carbohydrates are one of three macronutrients - along with proteins and fats - that the body requires daily. There are three main types of carbohydrates : starches, fiber, and sugars. Carbohydrates are a source of energy : They help fuel brain, kidneys, heart muscles, and central nervous system. For instance, fiber is a carbohydrate that aids in digestion, helps you feel full, and keeps blood cholesterol levels in check. Human's and animal's body can store extra carbohydrates in muscles and liver for further use. A carbohydrate-deficient diet may cause headaches, fatigue, weakness, difficulty concentrating, nausea, constipation, bad breath and vitamin and mineral deficiencies.

This polycopy is compounded of two parts. Part one consists of carbohydrate's structure and the second one is about the metabolism pathway. The manuscript is presented in English for the first 1st veterinarian and medecin students.

The purpose of this work is to introduce biochimistry module in simple, easy and understandable ways.

Carbohydrates. (Lehninger, Principles of Biochemistry, 4th ed; Berg, et al., 8Th Ed.

**I.Carbohydrates** (saccharides) structure : are molecular compounds made from three elements (carbon, hydrogen and oxygen). They are often called sugars (Glucose, sucrose). Polysaccharides are large carbohydrate molecules (such as starch and cellulose).

- They are a source of energy.
- Building blocks for polysaccharides (giant carbohydrates).
- Components of other molecules e.g. DNA, RNA, glycolipids, glycoproteins, ATP.

#### 1. Monosaccharides : (Lehninger, Principles of Biochemistry, 4th ed)

Monosaccharides are the simplest carbohydrates and are often called single **sugars**. They are the building blocks from which all bigger carbohydrates are made. They have the general molecular formula (CH2O)*n*.

They can be classified according to the number of carbon atoms in a molecule :

n = 3 Triose, n=4 : Tetrose, n=5 : Pentose and n=6 : Hexose.

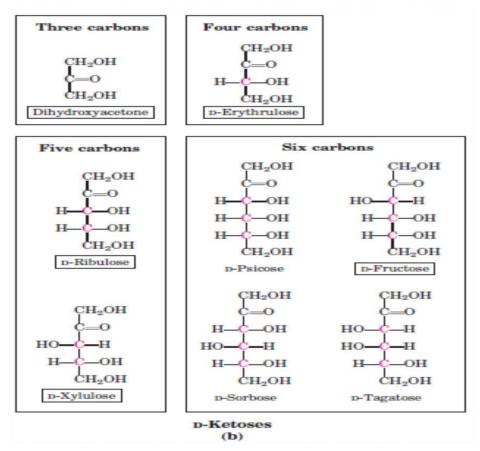


Figure 01 : Building blocks for polysaccharides

- 2. Isomerism : (Biochemistry, Berg, et al., 8Th Ed. (Stryer)
- **2.1.Isomers :** are compounds with the same molecular formula but having different structures or different orientation.

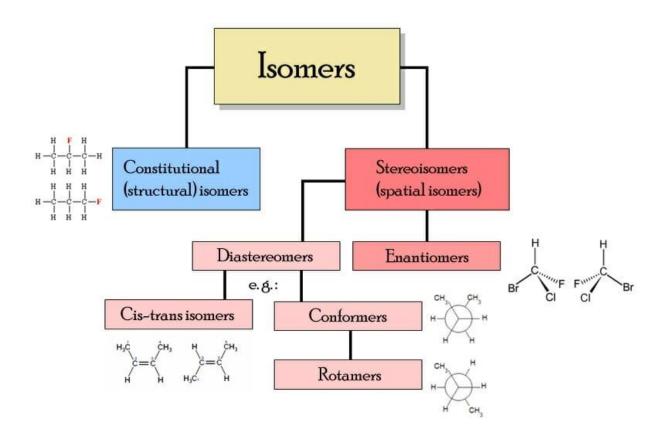


Figure 02 : Isomers structure. (https://www.google.com/search=isomers)

2.2. Structure (constitutional) isomers : They are two types :

- Functional group isomers : with different functional groups e.g : (aldehyde and ketone)

- **Positional isomers :** with substituent groups on different C-atoms e.g. (2-Phosphoglycerate and 3-Phosphoglycerate)

#### 2.3. Stereoisomers

Compounds with the same molecular formula, functional groups, and position of functional groups but have different conformations.

#### - optical isomers

Compounds with different conformation around chiral or asymmetric carbon atoms

The carbon C is asymmetric if A, B, D, and E are different groups

The four different groups A, B, D, and E can be arranged in space around the C-atom in two different ways to generate two different compounds

One member of an *enantiomeric pair* will rotate a plane of polarized light in a clockwise direction.(*dextrorotatory*) which is labelled (+)

The other member of the pair will rotate the light in a counterclockwise direction. ( *levorotatory*) which is labelled (-).

In general, a molecule with n chiral centers can have 2n stereoisomers.

e.g. Glyceraldehyde has 21 = 2;

Aldohexoses, with four chiral centers, have 24 = 16 stereoisomers.

**D**-Glyceraldehyde is the isomer that has the hydroxyl group on the **RIGHT** when the aldehyde group is at the top in a Fischer projection formula.

It is also dextrorotatory, so it is also  $\mathbf{D}(+)$ -Glyceraldehyde

L-Glyceraldehyde is the isomer that has the hydroxyl group on the **LEFT** when the aldehyde group is at the top in a Fischer projection formula.

It is also levorotatory, so it is also L(-)-Glyceraldehyde

#### 2.4. Enantiomers :

Enantioers are pairs of stereoisomers that are mirror images of each other.

#### 2.5. Diastereomers :

Optical isomers that are not enantiomers are *diastereomers* 

Diastereomers are pairs of stereoisomers that are not mirror images of each other.

#### 2.6. Epimers

Epimers are types of diastereomers that differ by their configuration on a single asymmetric carbon.

Ex.

-D-glucose and D-mannose, are epimers differ only in the stereochemistry at C-2.

- D-glucose and D- galactose are also epimers at C-4

**3. Monosaccharides Cyclic Structures.** (Lehninger, Principles of Biochemistry, 4th ed). In fact, in aqueous solution, aldotetroses and all monosaccharides with five or more carbon atoms in the backbone occur predominantly as cyclic (ring) structures in which the carbonyl group has formed a covalent bond with the oxygen of a hydroxyl group along the chain. The formation of these ring structures is the result of a general reaction between alcoholic OH and the carbonyl group of the aldehydes or ketones to form derivatives called hemiacetals or hemiketals.

#### 3.1. Anomers :

Are isomeric forms of the same monosaccharide that differ only in their configuration about the hemiacetal or hemiketal carbon atom.

- The hemiacetal carbon atom (or carbonyl) is called the **anomeric** carbon.

-D-glucose exists in solution as an intramolecular hemiacetal in which the free hydroxyl group at C-5 has reacted with the aldehydic C-1, rendering the latter carbon asymmetric and producing two anomers designated as  $\alpha$  and  $\beta$ .

*Mutarotation* is the interconversion of  $\alpha$ - and  $\beta$ - anomers when polarized light is passed through an aqueous solution of the monosaccharide to reach equilibrium.

#### (Haworth perspective formulas)( Lehe formulas).

The reaction between the aldehyde group at C-1 and the hydroxyl group at C-5 forms a hemiacetal linkage, producing either of two stereoisomers, the  $\alpha$  and  $\beta$  anomers, which differ

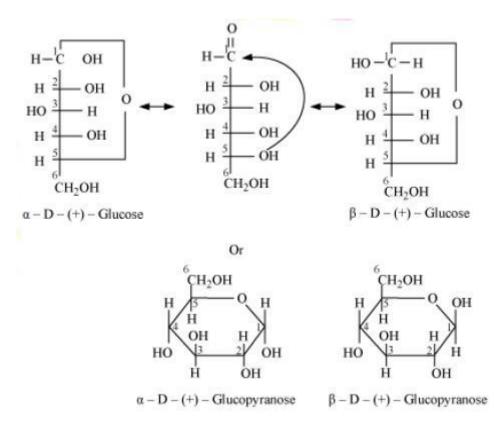
only in the stereochemistry around the hemiacetal carbon. These six-membered ring compounds are called pyranoses because they resemble the six membered ring compound pyran.

The systematic names for the two ring forms of D-glucose are  $\alpha$ -D-glucopyranose and  $\beta$ -Dglucopyranose.

Reaction between the ketone group at C-2 and the hydroxyl group at C-5 forms a hemiketal linkage, producing the  $\alpha$  and  $\beta$  anomers of fructose, which differ only in the stereochemistry around the hemiaketal carbon (C2). In Haworth perspective formulas of glucose and fructose, the edges of the ring nearest the reader are represented by bold lines.

#### 4. Monosaccharides structure.

#### 4.1.Glucose



#### Figure03 : Glucose structure (<u>https://www.google.com/search?q=glucose+structure</u>)

The main source of energy

- Binds to other sugars to give disaccharides, oligo and poly saccharides
- It is aldo hexose, glucopyranose
- It is called Dextrose
- (dextro-rotarory) +52.5
- It is reducing sugar

4.2.Fructose

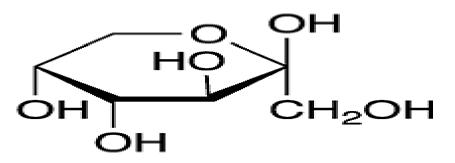
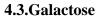


Figure 04 : Fructose structure. (<u>https://www.google.com/search?q=fructose+structure</u>)

- It is Keto hexose
- It is called levulose (levo rotatory) -92.3
- It is reducing sugar
- It is called also fruit sugar
- It binds to glucose to give sucrose
- Surprisingly, fructose form both pyranose and furanose rings.



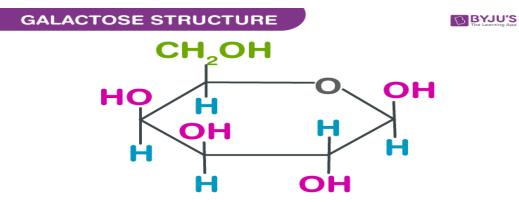


Figure 05 : Galactose structure. (<u>https://www.google.com/search?q=galactose+structure</u>)

Binds to glucose to give lactose

- It is aldo hexose
- It is reducing sugar
- It is epimer to glucose at C-4

• Galactose can be converted in the cell into glucose to be used for energy production.

#### 4.4.Ribose

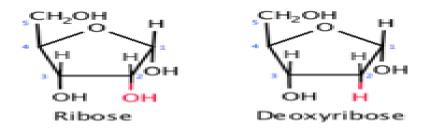


Figure 06 : Ribose structure. (https://www.google.com/search?q=ribose+structure)

The sugar in the nucleic acid

- Aldo pentose
- Forms part of some enzyme's cofactors.
- The DNA contains the deoxy form of ribose, lack of Oxygen in position C2
- The RNA contains the normal form of ribose
- It is reducing sugar

#### 5. Reactions of simple sugars.

They are different reaction

- Oxidation-reduction (required for monosaccharide metabolic breakdown)
- Esterification, Phosphorylation (reaction with alcohol, production of phosphate esters)
- Amino derivatives (used to produce structural components)
- Glycoside formation (linkage of monosaccharides to form oligo- and polysaccharides)

#### **5.1.Reducing**, nonreducing sugars.

- The hemiacetal linkage is weak and can be dissociated to give the open form.
- This process makes the carbonyl group available to reduce mild oxidizing agent such as ferric (Fe+3) or cupric (Cu+2) ion and the *carbonyl carbon* is oxidized to a *carboxyl group* plus ferrous (Fe+2) or cuprous (Cu+).
- This property is the basis of Fehling's reaction, a test for the presence of reducing sugar.

• By measuring the amount of oxidizing agent reduced by a solution of a sugar, we estimate the concentration of that sugar.

#### Highlight :

# Sugars with a free or potentially free aldehyde or ketone group (hemiacetal or hemiketal) have reducing properties and are called *reducing sugars*.

#### 5.2. Oxidation of aldoses (using Fehling reagent)

The presence of the hydrogen atom in the aldehyde group gives reducing properties to the aldehyde molecule. Aldehydes are easily oxidized by mild oxidizing agents such as Fehling's reagent to carboxylic acids Thus, aldoses are reducing agents.

Any sugar that has a free aldehyde group is referred to as a reducing sugar.

The product name is made by changing the –ose ending to –onic acid (-onate)

Glucose ------ gluconic acid or gluconate

Tests for reducing sugars: Fehling's reaction

#### **5.3.Oxidation of ketoses (using Fehling reagent)**

• Ketones do not have such reducing properties of the aldehyde group due to the lack of H attached to C=O and are not oxidized under similar conditions. fructose reduces such reagents even though it contains no aldehyde group.

#### **5.4.**Esterification

The most important biological esters of carbohydrates are phosphate esters. In the cell, phosphate esters are produced by transfer of phosphate group from ATP to carbohydrate hydroxyl group, a reaction catalyzed by enzymes called kinases.

#### 6. Oligosaccharides

#### **Disaccharides Contain a Glycosidic Bond**

Disaccharides (Maltose, lactose, and sucrose) consist of two monosaccharides joined covalently by an O-glycosidic bond, which is formed by dehydration (removal of H2O), a hydroxyl group of one sugar and hydrogen from OH of other sugar. This reaction form an acetal from a hemiacetal.

#### 6.1.Maltose

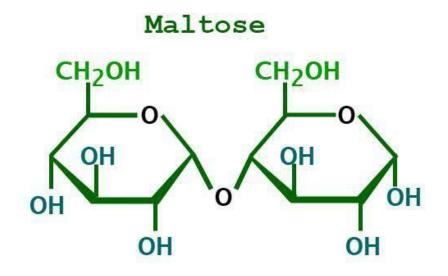


Figure 07 : Maltose structure. (<u>https://www.google.com/search?q=maltose+structure</u>)

It is also called maltobiose or malt sugar.

The disaccharide *maltose* contains two D-glucose residues joined by a O-glycosidic linkage between C-1 (the anomeric carbon) of one glucose residue and C-4 of the other. The configuration of the anomeric carbon atom in the glycosidic linkage is  $\alpha$ -(1-4).

6.2.Isomaltose

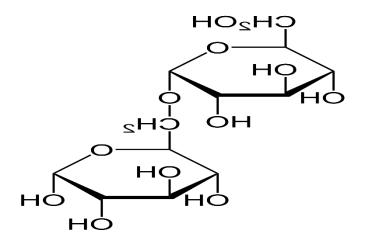
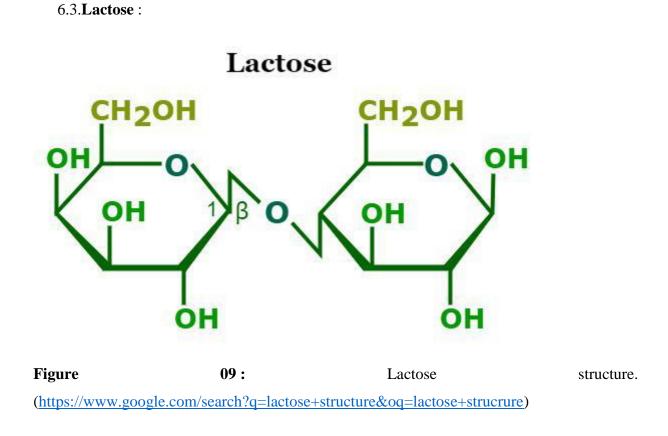


Figure 08 : Isoaltose structure. (<u>https://www.google.com/search?q=isomaltose+structure</u>).

Two units of glucose bound by  $\alpha$  (1-6) glycosidic bond. Isomaltose is a reducing sugar. It is produced by the hydrolysis of glycogen or amylopectin.



It is also called milk sugar.

It is composed of galactose and glucose linked by is  $\beta$ -(1-4) glycosidic bond.

lactose is a reducing disaccharide. Its abbreviated name is Gal(1-4)Glc.

#### **6.4.Sucrose : Saccharose**

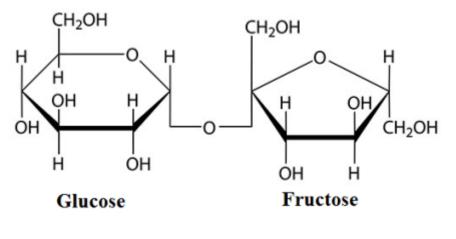


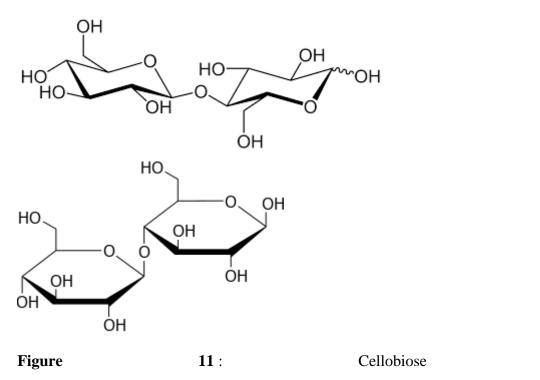
Figure10:Sucrosestructure.(https://www.google.com/search?q=sucrose+structure&oq=sucrose).

Sucrose (table sugar or cane sugar) is a disaccharide of glucose and fructose.

It is formed by plants but not by animals.

Sucrose contains no free anomeric carbon atom; the anomeric carbons of both monosaccharide units are involved in the glycosidic bond ( $\alpha 1$ ,  $\beta 2$ ). Sucrose is a non-reducing sugar.

#### 6.5 Cellobiose :



structure.

(https://www.google.com/search?q=cellobiose+structure&oq=cellobiose).

Two units of glucose bound by  $\beta$ -1-4 glycosidic bond. Cellobiose is a reducing sugar. It is produced by the hydrolysis of cellulose.

#### 7. Polysaccharides

Monosaccharides can undergo a series of condensation reactions, adding one unit after another to the chain until very large molecules (polysaccharides) are formed. This is called **condensation polymerisation**, and the building blocks are called **monomers**.

#### 7.1.Starch

Starch is often produced in plants as a way of storing energy. It exists in two forms: **amylose** and **amylopectin**. Both are made from  $\alpha$ -glucose. Amylose is an unbranched polymer of  $\alpha$ -glucose. The molecules coil into a helical structure. It forms a colloidal suspension in hot water. Amylopectin is a branched polymer of  $\alpha$ -glucose. It is completely insoluble in water.

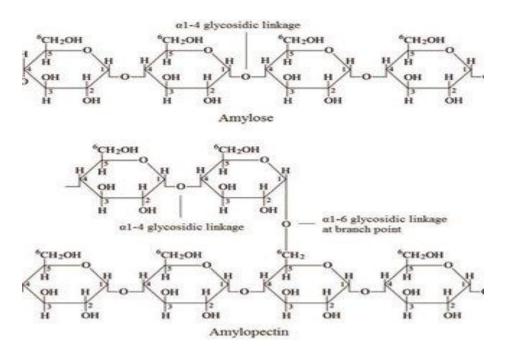
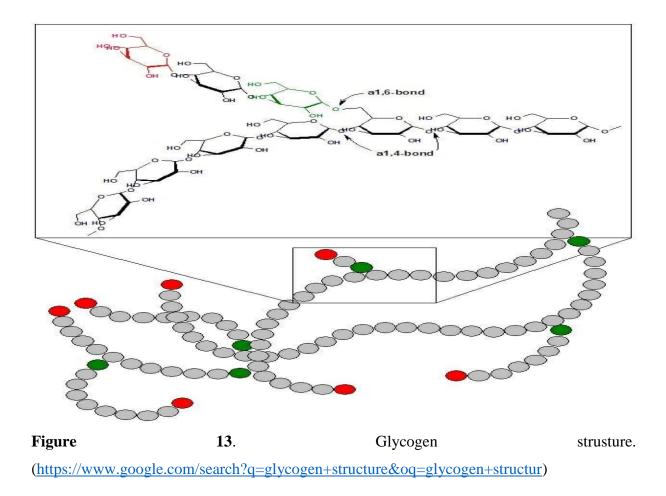


Figure 12 : Starch structure. (<u>https://www.google.com/search?q=starch+strucutre</u>)

#### 7.2.Glycogen

Glycogen is amylopectin with very short distances between the branching side-chains. Starch from plants is hydrolysed in the body to produce glucose. Glucose passes into the cell and is used in metabolism. Inside the cell, glucose can be polymerised to make glycogen which acts as a carbohydrate energy store.



#### 7.3.Cellulose

Cellulose is a third polymer made from glucose. But this time it's made from  $\beta$ -glucose molecules and the polymer molecules are 'straight'.

Cellulose serves a very different purpose in nature to starch and glycogen. It makes up the cell walls in plant cells. These are much tougher than cell membranes. This toughness is due to the arrangement of glucose units in the polymer chain and the **hydrogen-bonding** between neighbouring chains.

Cellulose is not hydrolysed easily and, therefore, cannot be digested so it is not a source of energy for humans. The stomachs of Herbivores contain a specific enzyme called cellulase which enables them to digest cellulose.

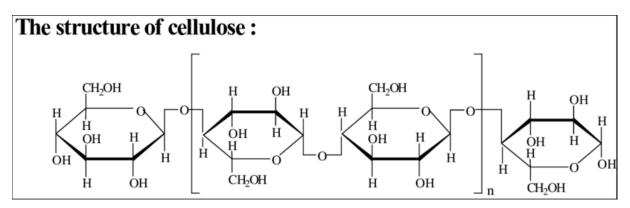
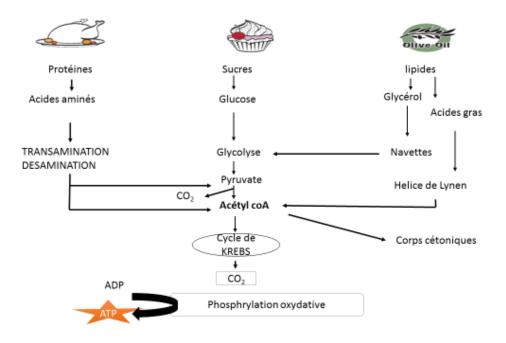


Figure 14 : Cellulose structure. (<u>https://www.google.com/search?q=cellulose+structure</u>)

**II. Metabolism.** (Harper's, 30E ., 2015 ; Chakrapani 3rd edition ; Lehninger 4ed edition ; Garrett And Grisham 2nd Ed 1998 ; Stryer 5th Edition repost ; Color Atlas of Biochemistry ., 2005).

Is the differnent chimical and enzymatique reactions in the cell –It devide into **Anabolism** and **Catabolism**.

A metabolic pathway is a series of enzyme-catalyzed reactions, so the pathway's substrate is converted to its product in discrete steps. A monomeric compound such as glucose is interconverted with its polymeric form (glycogen), with other monosaccharides (fructose-6-phosphate and ribose-5-phosphate, for example), and with smaller metabolites such as the three-carbon pyruvate.



**Figure 15 : Catabolism pathway** 

#### 1. Catabolism of Glucose (Glycolysis pathway)

Glucose occupies a central position in the metabolism of most cells. It is a major source of metabolic energy (in some cells, it is the only source), and it provides the precursors for the synthesis of other biomolecules. Recall that glucose is stored in polymeric form as starch in plants and as glycogen in animals.

The breakdown of these polymers provides glucose monomers that can be catabolized to release energy. This pathway of conversion of the six-carbon glucose to the three-carbon pyruvate, occurs in ten steps. This catabolic pathway is accompanied by the phosphorylation of two molecules of ADP (to produce 2 ATP) and the reduction of two molecules of NAD1. Reactions 1–5 are the energy-investment phase of glycolysis

The first phase requires the *investment* of free energy in the form of two ATP molecules.

#### 1.1. Hexokinase

In the first step of glycolysis, the enzyme hexokinase transfers a phosphoryl group from ATP to the C6 OH group of glucose to form glucose-6-phosphate: Hexokinase catalyzes a **metabolically irreversible reaction** that prevents glucose from backing out of glycolysis.

#### 1. 2. Phosphoglucose Isomerase

The second reaction of glycolysis is an isomerization reaction in which glucose-6- phosphate is converted to fructose-6-phosphate:

#### 1. 3. Phosphofructokinase

The third reaction of glycolysis consumes a second ATP molecule in the phosphorylation of fructose-6-phosphate to yield fructose-1,6-bisphosphate. CH2OH

Phosphofructokinase operates in much the same way as hexokinase, and the reaction it catalyzes is irreversible.

#### 1. 4. Aldolase

Reaction 4 converts the hexose fructose-1,6-bisphosphate to two three-carbon molecules, each of which bears a phosphate group.

This reaction is the reverse of an aldol (aldehyde–alcohol) condensation, so the enzyme that catalyzes the reaction is called aldolase.

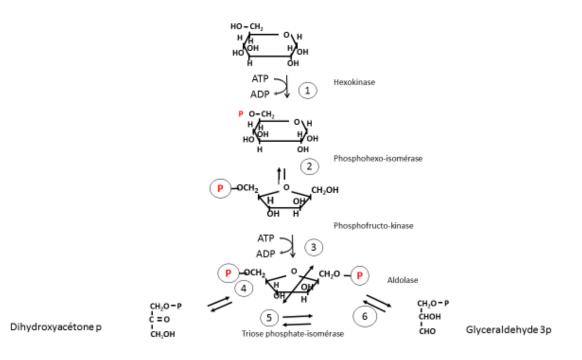


Figure 16 : Glycosis first phase.

#### 1. 5. Triose Phosphate Isomerase

The products of the aldolase reaction are both phosphorylated three-carbon compounds, but only one of them—glyceraldehyde-3-phosphate—proceeds through the remainder of the pathway. Dihydroxyacetone phosphate is converted to glyceraldehyde-3-phosphate by triose phosphate isomerase.

#### 1. 6. Glyceraldehyde-3-Phosphate Dehydrogenase.

In the sixth reaction of glycolysis, glyceraldehyde-3-phosphate is both oxidized and phosphorylated.

#### 1. 7. Phosphoglycerate Kinase

The product of Reaction 6, 1,3-bisphosphoglycerate, is an acyl phosphate.

#### 1. 8. Phosphoglycerate Mutase

In the next reaction, 3-phosphoglycerate is converted to 2-phosphoglycerate.

#### **Total energy**

• Glucose + 2 ADP + 2 Pi + 2 NAD<sup>+</sup> ----> 2 pyruvate + 2 ATP + 2 H<sub>2</sub>O + 2 NADH

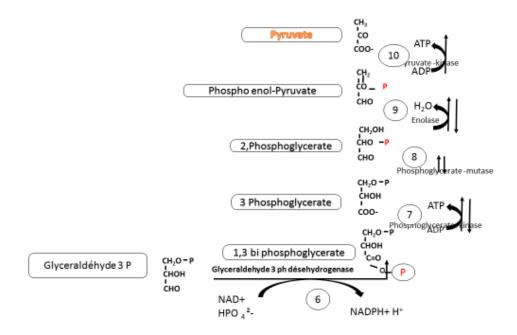


Figure 17 : Glycosis second phase.

2. Catabolism of Other Sugars

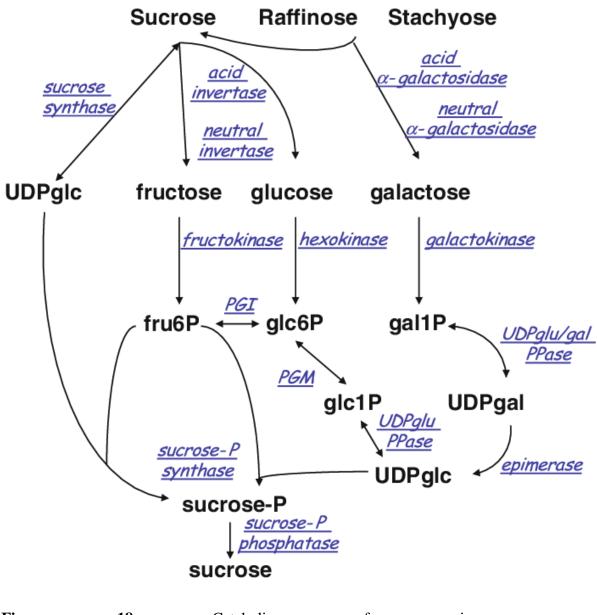
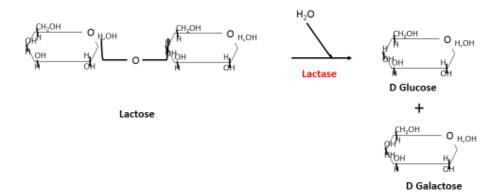


Figure18 :Catabolismofvarioussugars.(https://www.google.com/search?q=metabolism+of+various+sugars).

**2.1. Lactose :** is cleaved in the intestine by the enzyme lactase, and the two monosaccharides are absorbed, transported to the liver, and metabolized.

# Lactose( Diholoside du lait)



#### Figure 19 : Lactose catabolism.

2.2.**Sucrose :** is composed of glucose and fructose; sucrose is hydrolyzed in the small intestine, and its components glucose and fructose are absorbed

# Saccharose (Sucre de table)

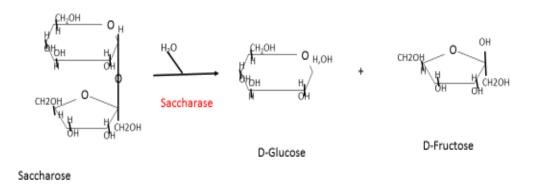


Figure 20 : Saccharose catabolism.

2.3.**Fructose** : is differs from the catabolism of glucose. Fructose is metabolized primarily by the liver, but the form of hexokinase present in the liver (called glucokinase) has very low affi nity for fructose. Fructose therefore enters glycolysis by a different route.

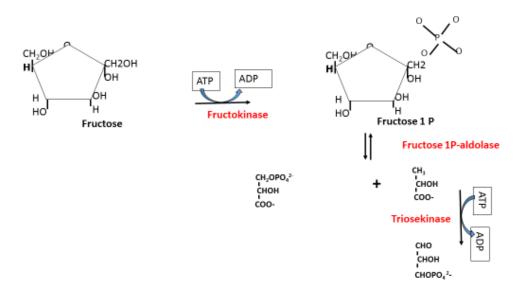


Figure 21 : Fructose catabolism.

#### 3. Pyruvate pathway :

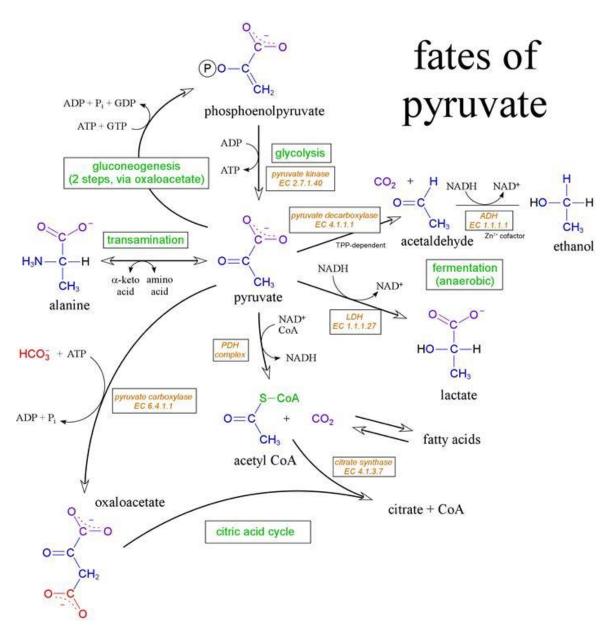
Pyruvate can be further broken down to acetyl-CoA or used to synthesize other compounds such as oxaloacetate.

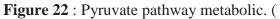
The fate of pyruvate depends on the cell type and the need for metabolic free energy and molecular building blocks.

During exercise, pyruvate may be temporarily converted to lactate. In a highly active muscle cell, glycolysis rapidly provides ATP to power muscle contraction, but the pathway also consumes NAD1 at the glyceraldehyde-3- phosphate dehydrogenase step.

Lactate represents a sort of metabolic dead end: Its only options are to be eventually converted back to pyruvate (the lactate dehydrogenase reaction is reversible) or to be exported from the cell. The liver takes up lactate, oxidizes it back to pyruvate, and then uses it for gluconeogenesis. The glucose produced in this manner may eventually make its way back to the muscle to help fuel continued muscle contraction. When the muscle is functioning aerobically, NADH produced by the glyceraldehyde-3-phosphate dehydrogenase reaction is reoxidized by oxygen and the lactate dehydrogenase reaction is not needed.

Organisms such as yeast growing under anaerobic conditions can regenerate NAD1 by producing alcohol. In the mid-1800s, Louis Pasteur called this process **fermentation**,. First, pyruvate decarboxylase (an enzyme not present in animals) catabolizes the removal of pyruvate's carboxylate group to produce acetaldehyde. Next, alcohol dehydrogenase reduces acetaldehyde to ethanol.





https://www.google.com/search?q=pyruvate+pathway+metabolic)

Ethanol is considered to be a waste product of sugar metabolism; its accumulation is toxic to other organisms (Box 13-B), including the yeast that produce it.

Although glycolysis is an oxidative pathway, its end product pyruvate is still a relatively reduced molecule. The further catabolism of pyruvate begins with its decarboxylation to form a two-carbon acetyl group linked to coenzyme A.

4. **Gluconeogenesis :** (Brosnan, J. T., Comments on metabolic needs for glucose and the role of gluconeogenesis, *Eur. J. Clin. Nutr.* **53**, S107–S111 (1999).

Pyruvate is converted to glucose by glycolytic enzymes operating in reverse and by enzymes that bypass the irreversible steps of glycolysis.

• Gluconeogenic flux is regulated primarily by fructose-2,6- bisphosphate.

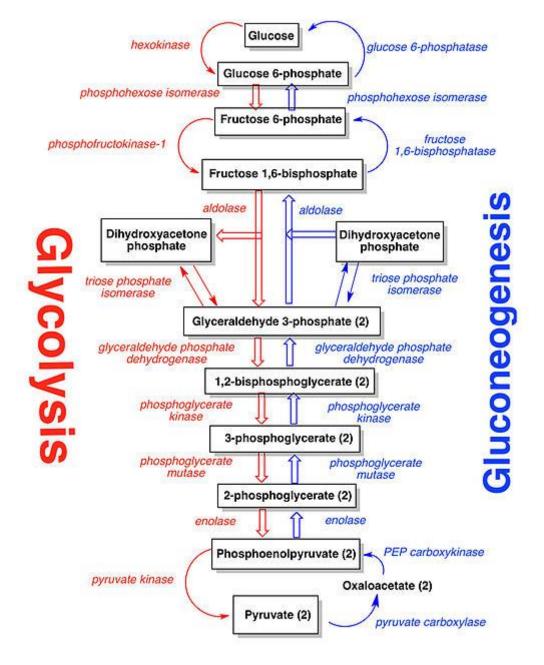


Figure23 :Gluconeogenosispathway.

(https://www.google.com/search?q=gluconeogenesis+pathway)

Pyruvate cannot be converted directly back to phosphoenolpyruvate because pyruvate kinase catalyzes an irreversible reaction (Reaction 10 of glycolysis). To get around this thermodynamic barrier, pyruvate is carboxylated by pyruvate carboxylase to yield.

Gluconeogenesis is energetically expensive. Producing 1 glucose from 2 pyruvate consumes 6 ATP, 2 each at the steps catalyzed by pyruvate carboxylase, phosphoenolpyruvate carboxykinase, and phosphoglycerate kinase. If glycolysis occurred simultaneously with gluconeogenesis, there would be a net consumption of ATP.

5. Glycogen Synthesis and Degradation : Greenberg, et *al* (2006). ; Özen, H., (2007).

Roach, et *al* (2012).

5.1.Glycogen degradation

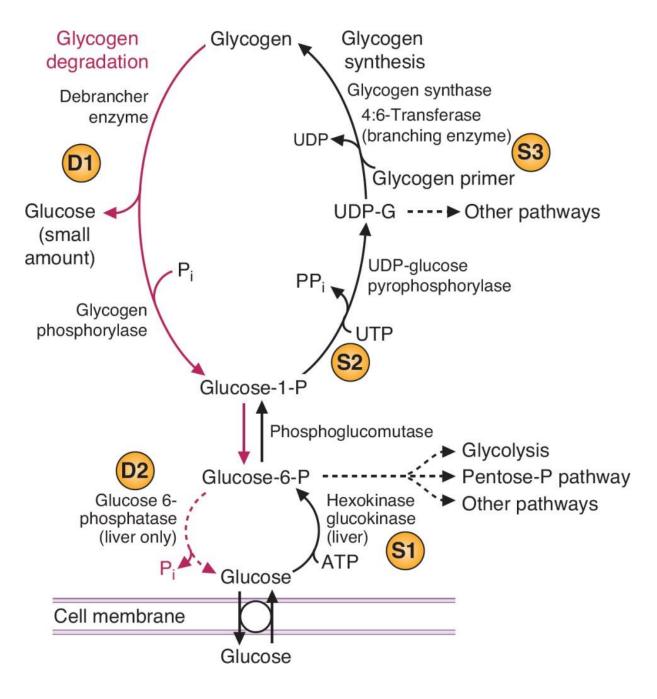


Figure24 :Starchandglycogendegradationpathway.(https://www.google.com/search?q=glycogen+degradation+pathway)

#### 5.2.Glycogen synthesis

The monosaccharide unit that is incorporated into glycogen is glucose-1-phosphate, which is produced from glucose-6-phosphate (the penultimate product of gluconeogenesis) by the action of the enzyme phosphoglucomutase.

 Pentose Phosphate Pathway: (Harper's, 30E ., 2015; Chakrapani 3rd edition; Lehninger 4ed edition; Garrett And Grisham 2nd Ed 1998; Stryer 5th Edition repost; Color Atlas of Biochemistry ., 2005).

The **pentose phosphate pathway**, which converts glucose-6-phosphate to ribose- 5-phosphate, is an oxidative pathway that occurs in all cells. But unlike glycolysis, the pentose phosphate pathway generates NADPH rather than NADH. The two cofactors are not interchangeable and are easily distinguished by degradative enzymes (which generally use NAD1) and biosynthetic enzymes (which generally use NADP1). The pentose phosphate pathway is by no means a minor feature of glucose metabolism. As much as 30% of glucose in the liver may be catabolized by the pentose phosphate pathway. This pathway can be divided into two phases: a series of oxidative reactions followed by a series of reversible interconversion reactions.

Pentose Phosphate pathway

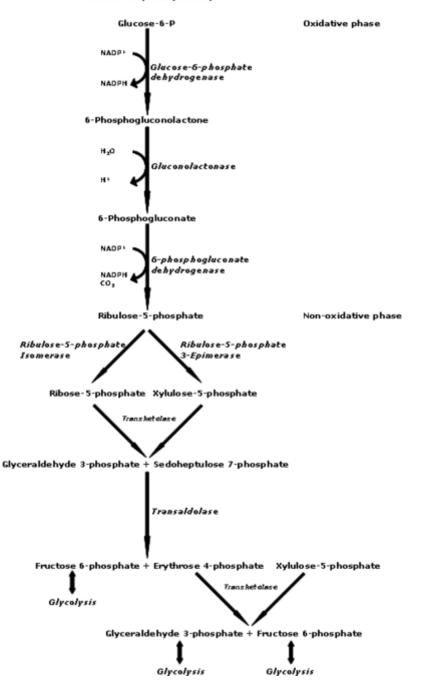


Figure 25 : Pentose pathway. (<u>https://www.google.com/search?q=pentose+pathway</u>)

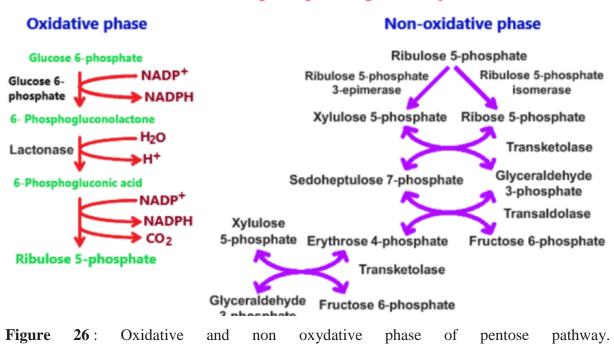
#### 6.1. Oxidative reactions of the pentose phosphate pathway.

The starting point of the pentose phosphate pathway is glucose-6-phosphate, which can be derived from free glucose, from the glucose-1-phosphate produced by glycogen phosphorolysis, or from gluconeogenesis. In the first step of the pathway, glucose-6-phosphate

dehydrogenase catalyzes the metabolically irreversible transfer of a hydride ion from glucose-6-phosphate to NADP1, forming a lactone and NADPH:

#### 6.2. Non oxydative of the pentose pathway.

The ribulose-5-phosphate product of the oxidative phase of the pentose phosphate pathway can isomerize to ribose-5-phosphate: Ribose-5-phosphate is the precursor of the ribose unit of nucleotides. In many cells, this marks the end of the pentose phosphate pathway, which has the net equation



## Pentose phosphate pathway

(<u>https://www.google.com/search?q=pentose+phosphate+pathway+oxygen</u>).

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