Carbohydrate metabolism

Introduction

Carbohydrates are an important source of energy for the human beings as well as a means by which chemical energy can be storage. Catabolism of carbohydrates provides the major share of the requirements for maintains of life.

Classification of carbohydrate:

Carbohydrate may be classified in three main groups

1. Monosaccharides: the simple sugar that have only four to seven or eight carbon atoms and contain only one aldehyde or ketone functional groups.
2. Disaccharide: contain two molecules of monosaccharide such as sucrose, lactose, and maltose
3. Oligosaccharide: compound sugars that yield two to six molecules of simple sugar on hydrolysis.
4. Polysaccharides: groups of compounds that yield a large number monosacchrides on hydrolysis (starch and glycogen).

Importance of Glucose:

1. Glucose is the preferred source of energy for most of the body tissues. Brain cells derive energy mainly from glucose.
2. When the glucose metabolism is deranged, life- threatening conditions may occur. A minimum amount of glucose is always required for normal functioning.
3. Normal fasting plasma glucose level is 70 to 110 mg/dl. After a heavy carbohydrate meal, in a normal person, this level is below 150 mg/dl.

Carbohydrate metabolism:

Sucrose, lactose, maltose, and isomaltose are the principle source of carbohydrate in the diet of the western world. Less than 3 % of the ingested carbohydrates is free glucose. Carbohydrate must be broken down to monosaccharides before they can be absorbed into the intestinal mucosal cells.

Sucrose ⟶ Fructose + Glucose

Lactose ⟶ Galactose + Glucose

Maltose ⟶ Glucose + Glucose

Disaccharide Monosacchride

In the digestive process, salivary amylase mix with food , hydrolyzed starch to starch dextrin, maltose and maltotriose. The digestion by the enzyme continuos until it inactivated by gastric HCl. Further hydrolysis occurs by the pancreatic amylase in the intestine.

Sucrose, lactose, maltose and isomaltose ingested in the diet or formed from amalase hydrolyzes, are further broken down into monosacchride by enzymes are attached to the brush border of the intestine cells. Monosacchrides are than transported to the liver via portal circulation.

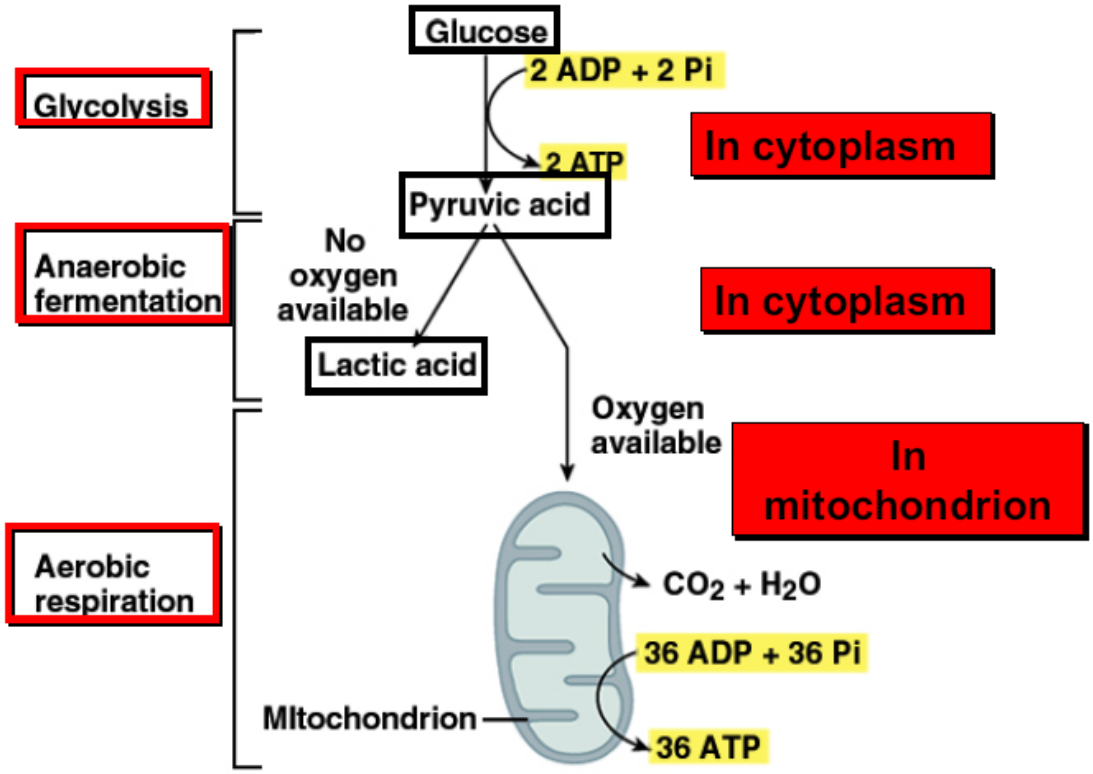
In the liver, fructose and galactose are mainly converted to glucose. Some glucose is released into circulating blood to supply energy to the tissues, or stored in form of glycogen in the liver and muscles. The catabolism of glucose of glucose to pyruvic acid or lactic acid for production of energy *via* ATP production is called **glycolysis.**

Glycoltic enzymes are present in the extra-mitochondrial compartment of the cell. Although aerobic glycolysis occurs in most tissues like liver, kidney, and erthrocytes, anaerobic glycolysis occures only in muscle.

Plasma glucose is derived from from the hydrolysis of the dietry starch and polysacared from the conversion of the other dietry hexose into glycose by the liver and the synthesis of glucose from the amino acids or pyruvate.

In times of glucose excess (elevation of blood glucose after the meal), glucose is enzematically polymerized in the liver to form glycogen (glycogenesis). When the glucose drops, glycogen is converted to glucose (glycogenolysis) by a defferent set of the enzymes thus, indepdent mechaanisms exits for regulation the blood glucose level by means of glycogenesis - glycogenolysis reactions.

The liver is the main orgain for the storage of excess carbohydrate as glycogen, although skeletal and the heart muscles can store minor amount. Excess glucose is converted to fat by the adipose cells where it stored.



The aerobic and anaerobic pathyways of carbohydrate metabolism

GLYCOLYSIS:

(Embden-Meyerhof Pathway) Importance of the Pathway

i) Glycolysis is derived from the Greek word, glykys = sweet; and lysis = splitting. In this pathway glucose is converted to pyruvate (aerobic condition) or lactate (anaerobic condition), along with production of a small quantity of energy.

ii) All the reaction steps take place in the cytoplasm. It is the only pathway that is taking place in all the cells of the body.

iii) Glycolysis is the only source of energy in erythrocytes.

iv) In strenuous exercise, when muscle tissue lacks enough oxygen, anaerobic glycolysis forms the major source of energy for muscles.

v) The glycolytic pathway may be considered as the preliminary step before complete oxidation.

vi) The glycolytic pathway also provides carbon skeletons for synthesis of certain non-essential amino acids as well as glycerol part of fat.

vii) Most of the reactions of the glycolytic pathway are reversible, which are also used for gluconeogenesis.

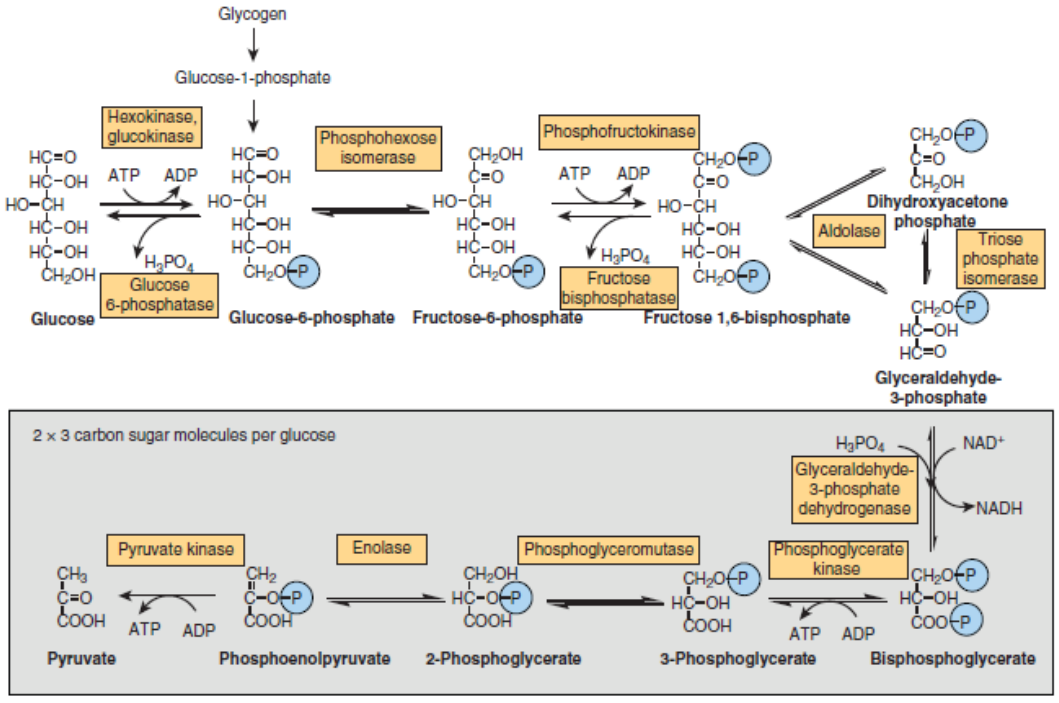


FIGURE . **The pathway of glycolysis.** (Ⓟ, —PO_3^{2-}; P_i $HOPO_3^{2-}$; ⊖, inhibition.) *Carbons 1-3 of fructose bisphosphate form dihydroxyacetone phosphate, and carbons 4-6 form glyceraldehyde-3-phosphate.

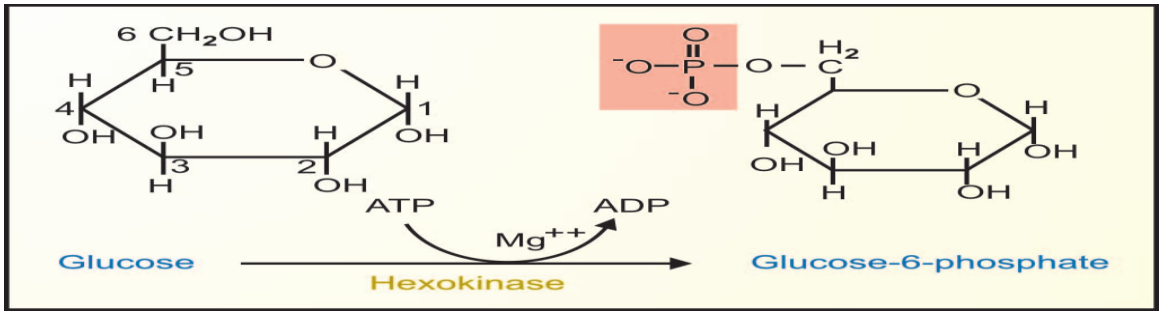
Summary of glycolysis (Embden-Meyerhof pathway). Steps 1, 3 and 9 are key enzymes; these reactions are irreversible. Steps 5, 6 and 9 produce energy.

Step 0: Glucose Entry into Cells Glucose transporter-4 (GluT4) transports glucose from extracellular fluid to muscle cells and adipocytes. This translocase is under the influence of insulin. So, in diabetes mellitus, insulin deficiency hinders the entry of glucose into the peripheral cells. But GluT2 is the transporter in liver cells; it is not under the control of insulin.

Step 1 of Glycolysis

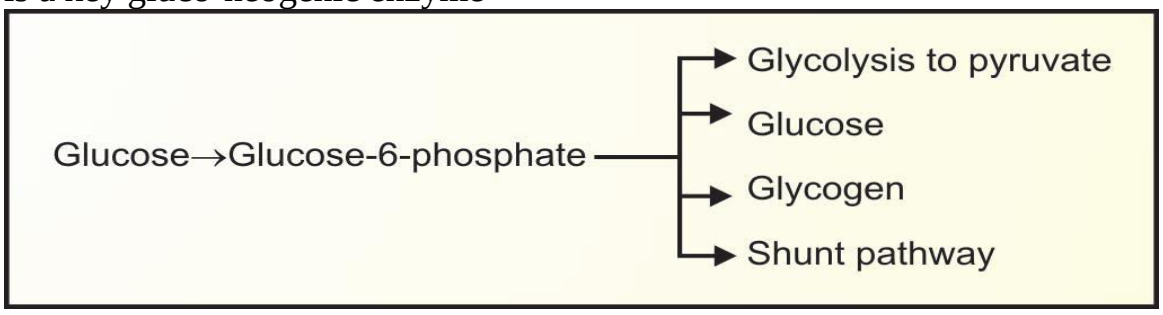
i) Glucose is activated by phosphorylation to glucose-6-phosphate .

ii) The enzyme is Hexokinase (HK), which splits the ATP into ADP, and the Pi is added on to the glucose. The energy released by the hydrolysis of ATP is utilised for the forward reaction.



iii) Hexokinase and glucokinase may be considered as iso-enzymes. Glucokinase is under the influence of insulin; but hexokinase is not.

iv) The metabolic fates of Glucose-6-phosphate are shown in plot.
v) The kinase reaction is irreversible; the same enzyme cannot produce glucose. But this irreversibility is circumvented by another enzyme glucose-6-phosphatase (see gluconeogenesis).
vi) Hexokinase is a key glycolytic enzyme, while glucose-6-phosphatase is a key gluco-neogenic enzyme



Step 2 of Glycolysis
Glucose-6-phosphate is isomerised to fructose-6-phosphate by an isomerase. This is readily reversible .
Step 3 of Glycolysis
i) Fructose-6-phosphate is further phosphorylated to fructose-1,6-bisphosphate . The enzyme is phospho fructo kinase (PFK).
ii) It needs ATP. PFK is an allosteric and inducible enzyme. It is an important key enzyme of this pathway.
iii) This reaction is an irreversible step in glycolysis. However this difficulty is circumvented by another enzyme, fructose-1,6-bisphosphatase.
Note:
The irreversible phosphorylation reaction catalyzed by phosphofructokinase-1 (PFK-1) is the most important control point and the rate-limiting and committed step of glycolysis. PFK-1 is controlled by the available concentrations of the substrates ATP and fructose 6-phosphate, and by regulatory substances described below. 1. Regulation by energy levels within the cell 2. Regulation by fructose 2,6-bisphosphate
Step 4 of glycolysis
The fructose-1,6-bisphosphate is cleaved into two halves; one molecule of glyceraldehyde-3-phosphate and another molecule of dihydroxyacetone phosphate. The enzyme is called aldolase. This reaction is reversible.
Step 4-A of Glycolysis
Dihydroxy acetone phosphate is isomerised to glyceraldehyde-3-phosphate by the enzyme triose phosphoisomerase. Thus net result is that glucose is now cleaved into 2 molecules of glyceraldehyde-3-phosphate.

For synthesis of neutral fat, glycerol is required which is derived from glucose through dihydroxy acetone phosphate.

Step 5 of glycolysis

In this step, Glyceraldehyde-3-phosphate is dehydrogenated and simultaneously phosphorylated to 1,3-bis-phospho glycerate (1,3-BPG) with
the help of NAD +. The enzyme is glyceraldehyde-3-phosphate dehydrogenase. This is a reversible reaction. The product contains a high energy bond.

Two major mechanisms for oxidizing NADH are:

1) The NADH-linked conversion of pyruvate to lactate (anaerobic), and
2) oxidation of NADH via the respiratory chain (aerobic).

Step 6 of Glycolysis

The energy of 1,3-BPG is trapped to synthesise one ATP molecule with the help of a phosphoglycerate kinase. This is an example of substrate level phosphorylation, where energy is trapped directly from the substrate, without the help of the complicated electron transport chain reactions (When energy is trapped by oxidation of reducing equivalents such as NADH, it is called oxidative phosphorylation). The reaction is reversible with the help of the same enzyme systems.

Step 7 of Glycolysis

3-phosphoglycerate is mutated to 2-phospho glycerate by shifting the phosphate group from 3rd to 2nd carbon atom. This is a readily reversible reaction.

Step 8 of Glycolysis

i) 2-phosphoglycerate is converted to phosphoenol pyruvate (PEP) by the enzyme enolase by the removal of a water molecule.
ii) The reaction is reversible.
iii) Enolase requires Mg ++ , and by removing these ions, fluoride will irreversibly inhibit this enzyme. By inhibiting enolase, fluoride will stop the whole glycolysis.
iv) So fluoride is added to blood; otherwise sugar is oxidised by the blood cells, so that when blood sugar is estimated after some time, false low values are obtained.

Step 9 of Glycolysis

i) Phospho enol pyruvate is dephosphorylated to pyruvate.
ii) The high energy content of PEP is trapped into ATP by the pyruvate kinase reaction. This is again an example of substrate level phosphorylation.

iii) The pyruvate kinase also is a key glycolytic enzyme. The pyruvate kinase step is irreversible. The reversal, however, can be attained in the body with the help of two enzymes and hydrolysis of 2 ATP molecules.
iv) While pyruvate kinase is a key glycolytic enzyme, the pyruvate carboxylase (PC) and phosphoenol pyruvate carboxy kinase (PEPCK) are key gluconeogenic enzymes.

Step 10 of Glycolysis

i) In anaerobic condition, pyruvate is reduced to lactate by lactate dehydrogenase (LDH) . (Anaerobiasis is a Greek term; a=not; aer=air; bios=life).
ii) LDH has 4 subunits and 5 iso-enzymes. The cardiac iso-enzyme of LDH has special importance to detect myocardial infarcts.
iii) When oxygen is available, the main metabolic fate of pyruvate is dehydrogenation to produce acetyl CoA; which then enters into citric acid cycle. However citric acid cycle can be operated only when there is plenty of oxygen. In the actively contracting muscles, there is comparative lack of oxygen. In such anaerobiosis, the major pathway of pyruvate is thus blocked.

Importance of phosphorylated intermediates:

1. **Possession of negative charge which inhibit their diffusion through membrane.**
2. **Conservation of free energy in high energy phosphate bond.**
3. **Facilitation of catalysis.**

Energy Yield from Glycolysis

1. During anaerobic (oxygen deficient) condition, when one molecule of glucose is converted to 2 molecules of lactate, there is a net yield of 2 molecules of ATP. On the whole, 4 molecules of ATP are synthesised by the 2 substrate level phosphorylations (steps 6 and 9). But 2 molecules of ATP are used in the steps 1 and 3, hence the net yield is only 2 ATP. The whole reaction is summarised as

Glucose + 2 Pi + 2 ADP 2 Lactate + 2 ATP

2. When oxygen is in plenty, the two NADH molecules, generated in the glyceraldehyde-3-phosphate dehydrogenase reaction (step 5), can enter the mitochondrial electron transport chain for complete oxidation. As each NADH provides 3 ATPs, this reaction generates 3 x 2 = 6 ATPs. Thus when oxygen is available, the net gain of energy from the glycolytic pathway is 8 ATPs (Table 5.2). Hence the ATP yield from glycolysis is different in anaerobic and aerobic conditions .

3. Complete oxidation of glucose: Pyruvate is later oxidatively decarboxylated to acetyl CoA, which enters into the citric acid cycle.

Formation of pyruvate producing ATP:

The conversion of PEP to pyruvate is catalyzed by pyruvate kinase, the third irreversible reaction of glycolysis. The equilibrium of the pyruvate kinase reaction favors the formation of ATP. [Note: This is another example of substrate-level phosphorylation.]

1. Feed-forward regulation: In liver, pyruvate kinase is activated by fructose 1,6-bisphosphate, the product of the phosphofructokinase reaction. This feed-forward (instead of the more usual feedback) regulation has the effect of linking the two kinase activities: increased phosphofructokinase activity results in elevated levels of fructose 1,6-bisphosphate, which activates pyruvate kinase.

2. Covalent modulation of pyruvate kinase: Phosphorylation by a cAMP-dependent protein kinase leads to inactivation of pyruvate kinase in the liver

3. Pyruvate kinase deficiency: The normal, mature erythrocyte lacks mitochondria and is, therefore, completely dependent on glycolysis for production of ATP. This high-energy compound is required to meet the metabolic needs of the red blood cell, and also to fuel the pumps necessary for the maintenance of the biconcave, flexible shape of the cell, which allows it to squeeze through narrow capillaries. The anemia observed in glycolytic enzyme deficiencies is a consequence of the reduced rate of glycolysis, leading to decreased ATP production.

Regulation of Glycolysis

1. Hormones

i) Insulin favours glycolysis by activating key glycolytic enzymes (glucokinase, phospho-fructokinase and pyruvate kinase).

ii) Glucagon and glucocorticoids inhibit glycolysis.

iii) Glucocorticoids inhibit glycolysis and favours gluconeogenesis.

2. PhosphoFructo Kinase (PFK) It is the most important rate-limiting enzyme for glycolysis pathway. PFK (step 3) is an allosterically regulated enzyme. ATP is the most important allosteric inhibitor. Yet another allosteric inhibitor of PFK is citrate. AMP acts as an allosteric activator.

3. Hexokinase and Glucokinase Glucose-6-phosphate inhibits hexokinase. But glucokinase is not affected by glucose-6-phosphate.

4. Pyruvate Kinase Pyruvate kinase catalyses an irreversible step (step 9). It is a regulatory enzyme of glycolysis. The enzyme is inhibited by ATP. F-1,6-BP on the other hand, activates pyruvate kinase. Insulin increases its activity whereas glucagon inhibits.

Reduction of pyruvate to lactate:

Lactate, formed by the action of lactate dehydrogenase, is the final product of anaerobic glycolysis in eukaryotic cells. The formation of lactate is the major fate for pyruvate in lens and cornea of the eye, kidney medulla, testes, leukocytes and red blood cells, because these are all poorly vascularized and/or lack mitochondria.

1. Lactate formation in muscle: In exercising skeletal muscle, NADH production (by glyceraldehyde 3-phosphate dehydrogenase and by the three NAD+-linked dehydrogenases of the citric acid cycle, exceeds the oxidative capacity of the respiratory chain.

2. Lactate consumption: The direction of the lactate dehydrogenase reaction depends on the relative intracellular concentrations of pyruvate and lactate, and on the ratio of NADH/NAD+ in the cell. For example, in liver and heart, the ratio of NADH/NAD+ is lower than in exercising muscle. These tissues oxidize lactate (obtained from the blood) to pyruvate. In the liver, pyruvate is either converted to glucose by gluconeogenesis or oxidized in the TCA cycle. Heart muscle exclusively oxidizes lactate to CO2 and H2O via the citric acid cycle.

3. Lactic acidosis: Elevated concentrations of lactate in the plasma, termed lactic acidosis, occur when there is a collapse of the circulatory system, such as in myocardial infarction, pulmonary embolism, and uncontrolled hemorrhage, or when an individual is in shock.

Mechanism of arsenic poisoning: The toxicity of arsenic is explained primarily by the inhibition of enzymes such as pyruvate dehydrogenase, which require lipoic acid as a coenzyme . However, pentavalent arsenic (arsenate) also can prevent net ATP and NADH production by glycolysis, without inhibiting the pathway itself. The poison does so by competing with inorganic phosphate as a substrate for glyceraldehyde 3-phosphate dehydrogenase, forming a complex that spontaneously hydrolyzes to form 3-phosphoglycerate .

Synthesis of 2,3-bisphosphoglycerate (2,3-BPG) in red blood cells: Some of the 1,3-BPG is converted to 2,3-BPG by the action of bisphosphoglycerate mutase . 2,3-BPG, which is found in only trace amounts in most cells, is present at high concentration in red blood cells (increases O2 delivery).

2,3-BPG is hydrolyzed by a phosphatase to 3-phosphoglycerate, which is also an intermediate in glycolysis .