Basic Biochemistry

Carbohydrates lecture 3

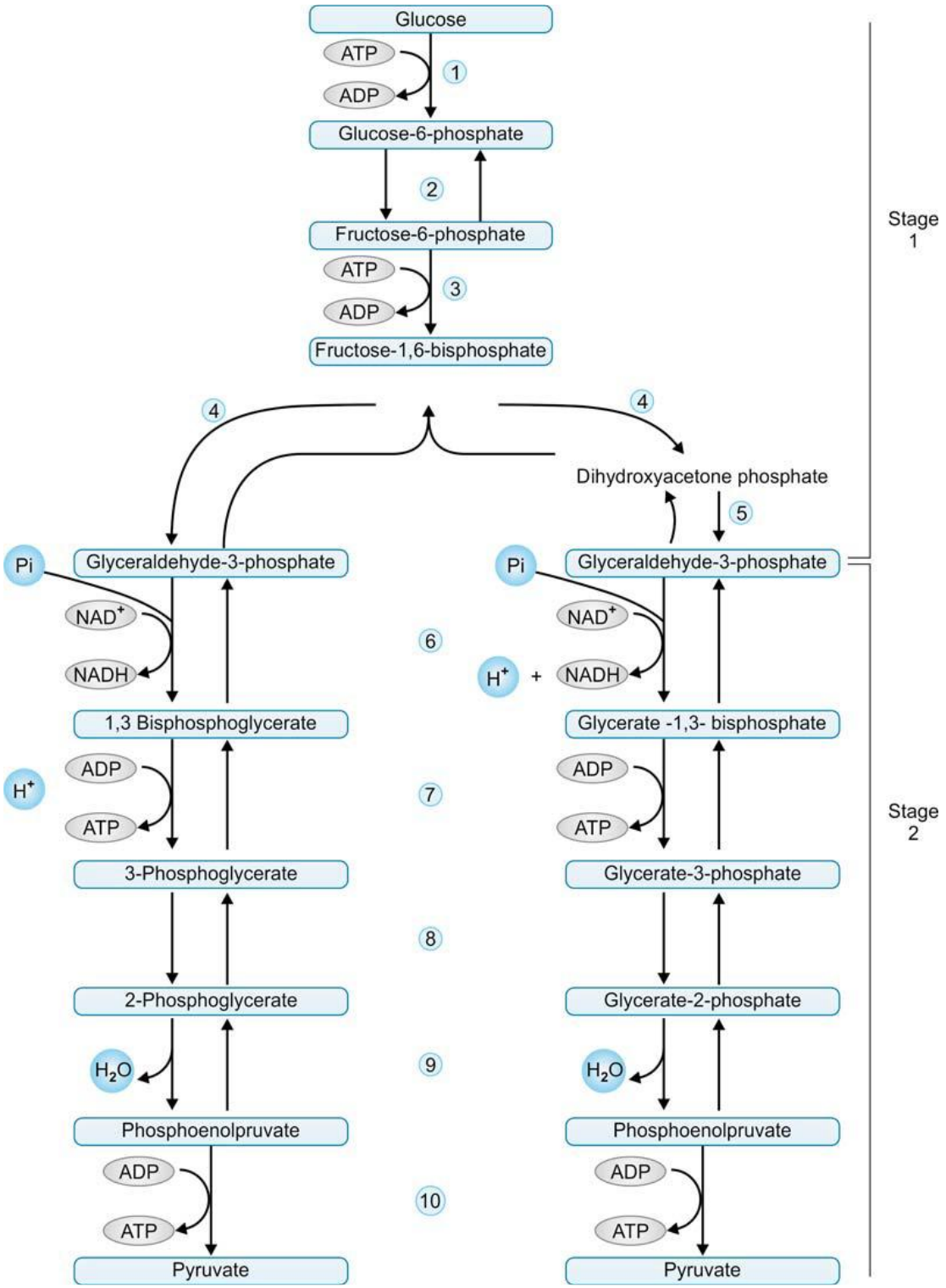
Dr. Dalya Shakir Obaida/ PhD Clinical Chemistry

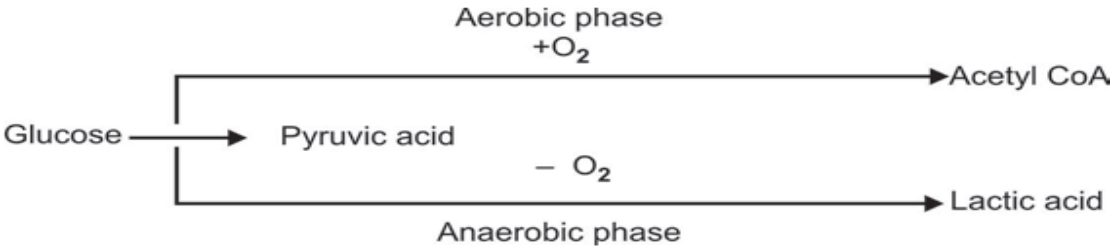
Metabolism of Carbohydrates

All the dietary carbohydrates are digested and absorbed as glucose, which enters the liver. In the liver, glucose is utilized in various metabolic pathways which include glycolysis (oxidized to produce energy), glycogenesis (stored as glycogen in the liver and muscle) and the pentose phosphate pathway (to produce pentoses and NADPH). During starvation, when blood glucose is low, glucose is released from glycogen (glycogenolysis) or produced via gluconeogenesis.

GLYCOLYSIS

The breakdown of glucose to pyruvic acid is called glycolysis. Under aerobic condition, pyruvic acid enters mitochondria and is completely oxidized to CO2 and H2O. Whereas, under anaerobic conditions, pyruvate is converted to lactic acid. The sequence of reactions from glucose to pyruvic acid is also called the Embden-Meyerhof pathway. Glucose is converted to pyruvate in 10 steps by glycolysis. Glycolysis is an extramitochondrial pathway and is carried out by a group of eleven enzymes.


Glucose
ATP
ADP
1
Glucose-6-phosphate
2
Fructose-6-phosphate
ATP
ADP
3
Fructose-1,6-bisphosphate
4
4
Dihydroxyacetone phosphate
5
Stage 1
Pi
Glyceraldehyde-3-phosphate
Pi
Glyceraldehyde-3-phosphate
NAD+
NADH
6
H+
+
NAD+
NADH
1,3 Bisphosphoglycerate
Glycerate -1,3- bisphosphate
ADP
ATP
H+
7
ADP
ATP
Stage 2
3-Phosphoglycerate
Glycerate-3-phosphate
8
2-Phosphoglycerate
Glycerate-2-phosphate
H2O
9
H2O
Phosphoenolpruvate
Phosphoenolpruvate
ADP
ATP
10
ADP
ATP
Pyruvate
Pyruvate



Salient Features of Embden-Meyerhof Pathway

1. The rate-limiting step in glycolysis is phosphofructokinase (PFK). PFK is stimulated by fructose-6-phosphate, AMP and ADP but is inhibited by ATP and citrate.

2. All the reactions of glycolysis are reversible except hexokinase, phosphofructokinase and pyruvate kinase catalyzed reactions because of energy barriers.

Glycolysis has three principal features:

1. It is the degradative pathway whereby D-glucose is oxidized to pyruvate, which is further metabolized by either of the two routes.
   A. When the supply of oxygen is inadequate for complete oxidation, the pyruvate is reduced to lactate.
   B. When the supply of oxygen is adequate (aerobic conditions)

the pyruvate is oxidatively decarboxylated to acetylCoA, which enters the citric acid cycle, where it is oxidized to carbon dioxide and water.

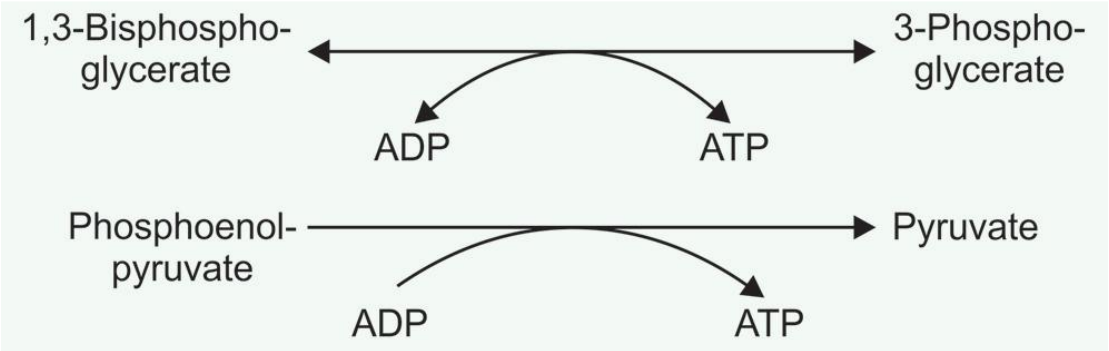
Bioenergetics (ATP production) of Glycolysis

Number of ATP produced during glycolysis vary, depending upon the condition.

A. **Under anaerobic condition**, conversion of 1,3bisphosphoglycerateto 3-phosphoglycerate and phosphoenolpyruvate to pyruvate, each produces one ATP, a total of 2 ATP is produced per molecule of a triose, or 4 molecules of ATP per molecule of hexose. As 2 ATP are used in the initial two reactions of the process, i.e. in the conversion of glucose to glucose- 6-phosphate and fructose-6-phosphate to fructose-1,6-

bisphosphate, the net energy yield per molecule of glucose is only 2 ATP.

B. **Under aerobic conditions** NADH, which is produced during the conversion of 3-phosphoglycerate to 1,3- bisphosphoglycerate can enter the electron transport chain and release 2.5 ATP, thus, additionally, 5 more ATP are produced per molecule of glucose. Therefore, a total yield under aerobic conditions is 9 ATP while the net yield is 7 ATP.



Biomedical Importance of Glycolysis

In the liver and other tissues, anaerobic glycolysis is the main source of ATP. In the muscle, anaerobic glycolysis provides energy during exercise.

Regulation of Glycolysis

There are three irreversible steps which are catalyzed by hexokinase, phosphofructokinase and pyruvate kinase. These are also the sites of the regulation of glycolysis.

Insulin stimulates these enzymes and increases utilization of glucose by glycolysis. On the other hand, **glucagon inhibits** this process.

TRICARBOXYLIC ACID CYCLE

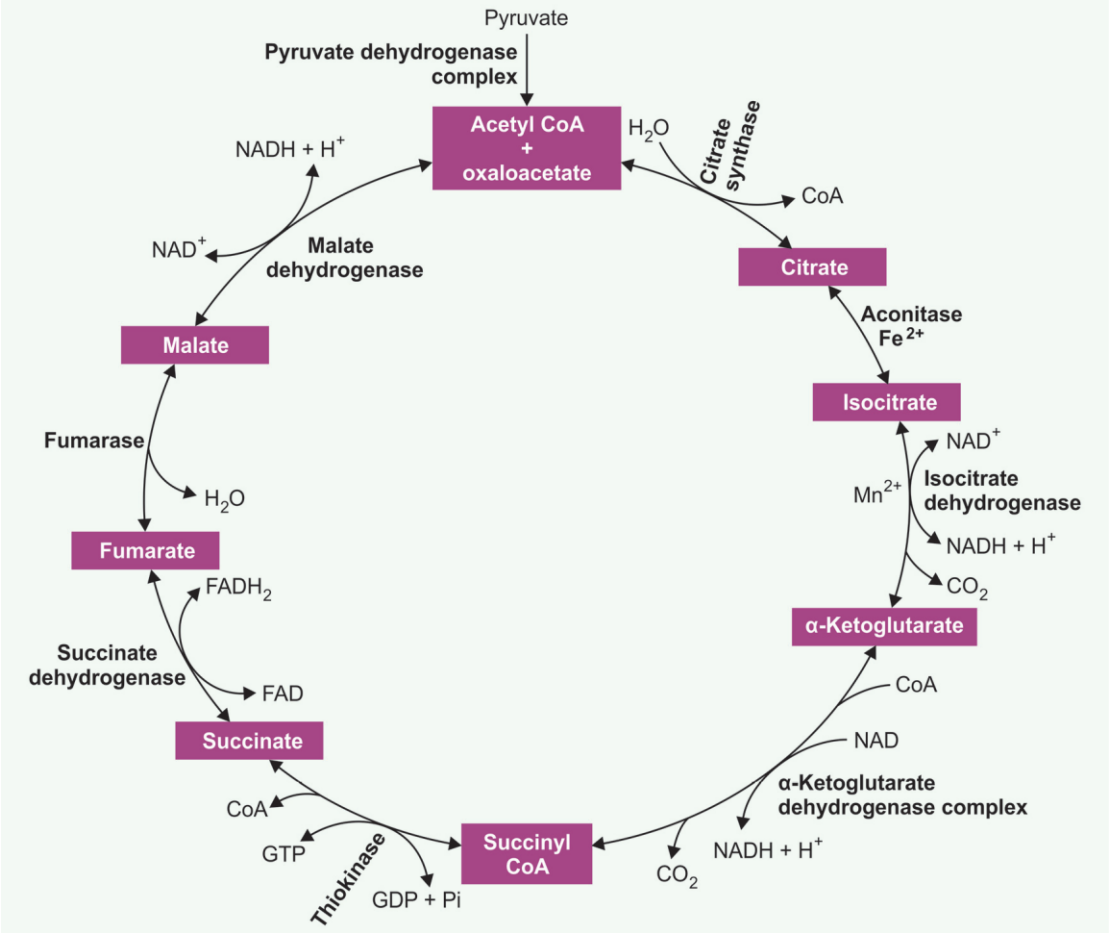
Tricarboxylic acid cycle (TCA cycle), also called **citric acid cycle** or **Krebs cycle**, is a process of the **oxidation of acetyl CoA** (active acetate, produced under aerobic glycolysis) to **CO2** and **H2O**. During oxidation of acetyl CoA reducing equivalents are produced, which enter respiratory chain and generate a large amount of ATP.

Outline of the TCA Cycle

Reactions of the TCA cycle are outlined in

1. In the first step, **acetyl-CoA** (formed from pyruvate under aerobic condition) combines with **oxaloacetate** and forms citric acid (a tricarboxylic acid). This reaction is catalyzed by the enzyme **citrate synthase**, also called the **condensing enzyme**.
2. **Citrate** is then rearranged to **cis-aconitate**, which is subsequently changed to isocitrate. Both of these steps are catalyzed by **aconitase**. Conversion of citrate to isocitrate is **inhibited by fluoroacetate**.
3. In the next step, in the presence of **isocitrate dehydrogenase**, **isocitrate** is converted to oxalosuccinate**,** which is subsequently decarboxylated to α-ketoglutarate.
4. Thereafter, α-ketoglutarate undergoes oxidative decarboxylation and gets converted to **succinyl CoA**. This reaction is similar to the conversion of pyruvate to acetyl CoA. **α-Ketoglutarate dehydrogenase complex**, which also requires five coenzymes, i.e. TPP, NAD+, FAD, coenzyme A and lipoic acid, catalyzes this reaction.
5. In the next step, **succinyl CoA** is converted to **succinate**, by the enzyme **succinate thiokinase**. During this reaction, a molecule of GTP is formed. This is known as **substrate level phosphorylation, as a high-energy** molecule is formed at the substrate level.

6. Thereafter, **succinate** is converted to **fumarate** by The enzyme **succinate dehydrogenase**. Due to structural similarities between malonate and succinate, **malonate inhibits** succinate dehydrogenase, **competitively**.
7. In the next step, with the addition of a molecule of water, by the enzyme **fumarase** (fumarate hydratase), **fumarate** is converted to **L-malate**.
8. Finally, **malate dehydrogenase**, in the presence of NAD+, converts **malate** to **oxaloacetate**.



Bioenergetics (ATP Production) of TCA Cycle

As a result of oxidation of one molecule of acetyl CoA in the Krebs cycle, three molecules of NAD+ and one molecule of FAD are reduced.

- Reducing equivalents (from NADH + H+) enter the respiratory chain at three different levels and result inthe production of 2.5 ATP at each level.
- FADH2 yields 1.5 ATP. Besides, there is also substrate

level production of GTP. Thus, **total ATP yield**, per molecule of acetyl CoA, is **10 ATP**

Reaction	Reducing equivalents produced as	Number of ATP produced
Isocitrate → α-Ketoglutarate	$NADH + H^+$	2.5
α-Ketoglutarate → Succinyl CoA	$NADH + H^+$	2.5
Succinyl CoA → Succinate	—	1
Succinate → Fumarate	$FADH_2$	1.5
Malate → Oxaloacetate	$NADH + H^+$	2.5
	Total	**10**

- For each molecule of glucose, 2 pyruvate molecules are formed. These are converted to 2 acetyl-CoAs, each of which is broken-down to 3 NADH, 1FADH2 and 1 GTP. Hence, for 1 glucose molecule, 6 NADH, 2 FADH2 and 2 GTP are produced in the TCA cycle.
- In glycolysis:

1. Reactions Where ATP is Consumed

- Glucose to glucose-6-phosphate 1 ATP
- Fructose-6-phosphate to fructose-1, 6-diphosphate 1 ATP

2. Reactions Where ATP is Generated

- Glyceraldehyde-3-PO4 to 1,3 diphosphoglycerate $2 \times 3 = 6$
- 1,3 diphosphoglycerate to 3-diphosphoglycerate $2 \times 1 = 2$
- Phosphoenolpyruvate to pyruvate $2 \times 1 = 2$

The ATP yield is 2

(Two molecules of ATP are generated in the conversion of glucose to pyruvate because NADH obtained in the glyceraldehyde-3-phosphate dehydrogenase reaction is not oxidized in mitochondria by the respiratory chain).

Under Aerobic Condition	**ATP**
• Pyruvate to acetyl CoA	2 × 3 = 6
• Isocitrate to oxalosuccinate	2 × 3 = 6
• α-ketoglutarate to succinyl CoA	2 × 3 = 6
• Succinyl CoA to succinate	2 × 1 = 2
• Succinate to fumarate	2 × 2 = 4
• Malate to oxaloacetate	2 × 3 = 6
	Total = 30

Total number of ATP molecules formed under aerobic conditions is 38, i.e. 30 from citric acid cycle and 8 from glycolysis.

Biomedical Importance of TCA Cycle

Citric acid cycle has a **dual role**, i.e. it is important in oxidation as well as synthetic processes. It is, thus, **amphibolic** in nature.

1) It is **catabolic** for the oxidation of carbohydrates, lipids and proteins, as they are completely oxidized to CO2 and H2O, and release energy.
2) It is also important in the **anabolic** reactions, as various intermediates of the cycle can be used for the biosynthesis of the nonessential amino acids.
3) Various intermediates of the cycle are also potentially glucogenic and thus, can give rise to glucose in liver and kidney.

dRegulation of TCA Cycle

TCA cycle is regulated by:

1) The availability of acetyl CoA,
2) Oxaloacetate and NADH.
3) Citrate synthase, isocitrate dehydrogenase and α-ketoglutarate dehydrogenase are the rate limiting enzymes of the TCA cycle.