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Biochemistry

carbohydratesisomersketone starchlipidproteinamine

Doctor 2017 | Medicine | JU

◉ Sheet ○ Slides

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NO NEED

Doctor

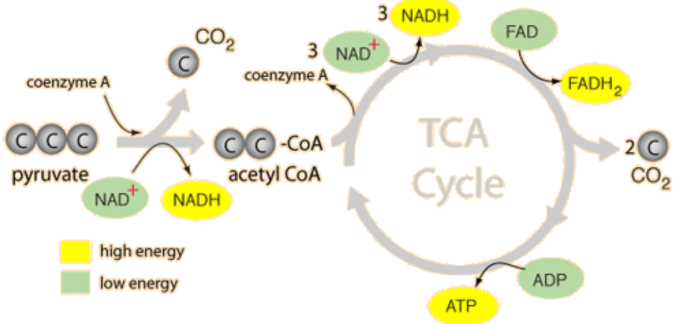
Nafez abu taroush

An introduction

Tricarboxylic Acid cycle (TCA cycle) is also called CITRIC Acid cycle or kreb`s cycle, it takes place in **mitochondrial matrix** and plays several roles in metabolism. This cycle shouldn't be viewed as a closed system but, instead, as an open one with compounds entering and leaving as well.

The TCA cycle functions as a metabolic pathway that further oxidizes organic fuel derived from pyruvate.

Pyruvate (final product of glycolysis) is broken down into 2-carbon units molecule called **Acetyl CoA.** Other sources from which we can get Acy.CoA is the oxidation of fatty acids and carbon degradation of amino acids.



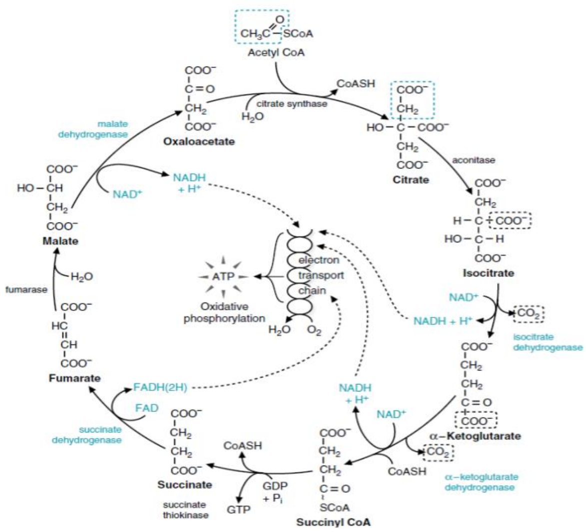
The cycle generates 1 ATP (or GTP) per turn by substrate-level phosphorylation, but most of the chemical energy is transferred to NAD^+ and FAD during redox reactions. The reduced coenzymes, NADH and $FADH_2$, shuttle their cargo of high-energy electrons into Electron Transport Chain (ETC).

Note that this step releases coenzyme A. The reaction is catalyzed by *citrate synthase*. The cycle has eight steps, each catalyzed by a specific enzyme. You can see that each turn of the cycle 2-carbon units molecule (Acetyl CoA) enter and two different carbons leave in the completely oxidized form of CO_2 molecules, and that makes sense!!! That's why it's called a cycle.

> NOTE **The first 4 steps are engaged in converting the 6-carbon units molecule into a 4-caron units molecule, while the other steps are engaged with reformulating (modifying) the remaining molecules.**

Step 1

The acetyl group of acetyl CoA joins the cycle by combining with the compound oxaloacetate, forming citrate. The formation of Citrate needs Energy, this Energy is obtained from the detachment of CoA. Note that this step releases coenzyme A and the reaction is catalyzed by *citrate synthase* (easy to memorize as the name implies its action). Citrate is the ionized form of citric acid, for which the cycle is named. The next seven steps decompose the citrate back



to oxaloacetate. It is this regeneration of oxaloacetate that makes the process a *cycle*.

NOTE **The principle of making Energy is all about oxidation reactions, catabolism is usually an oxidation process involving a release of chemical free energy. While anabolism usually involves reduction reactions.**

But Citrate molecule is considered as an exception since it's 3° alcohol as well as it has 3 carboxylic groups (highest oxidized form), thus it can't be oxidized. So, it must be converted to a different structure that is able to be oxidized, and the easiest way to do that is isomerization.

Step 2

Aconitase catalyzes the isomerization of citrate to isocitrate by hydroxyl group migration. In this reaction, a tertiary alcohol, which cannot be oxidized, is converted to a secondary alcohol.
Aconitase is so named because it goes through intermediates.

Step 3

Isocitrate then undergoes a reaction known as oxidative decarboxylation because the alcohol is oxidized, and the molecule is shortened by one carbon atom with the release of carbon dioxide (decarboxylation). The reaction is catalyzed by *isocitrate dehydrogenase*, and the product of the reaction is α-ketoglutarate (an acidic compound with 5-carbon units).
An important reaction linked to this is the reduction of the coenzyme nicotinamide adenine dinucleotide (NAD^+) to NADH. The NADH is ultimately reoxidized by ETC, and the energy released is used in the synthesis of ATP in a process called oxidative phosphorylation.

NOTE **The most common acceptor of e^- with dehydrogenases is NAD^+ (accepts a hydride). unless you have been informed with any other pieces of information.**

Step 4

it is another oxidative decarboxylation. This time α-ketoglutarate is converted to succinyl-CoA, and another molecule of NAD^+ is reduced to NADH. The *α-ketoglutarate dehydrogenase complex (α-KGDH)* catalyzes this reaction.
α-KGDH is a complex enzyme consisting of multiple copies of three subunits a thiamine pyrophosphate-dependent decarboxylase (E1), a trans acylase (lipoate) (E2), which uses lipoamide and coenzyme A (AKA CoASH), and a dehydrogenase (FAD) (E3).

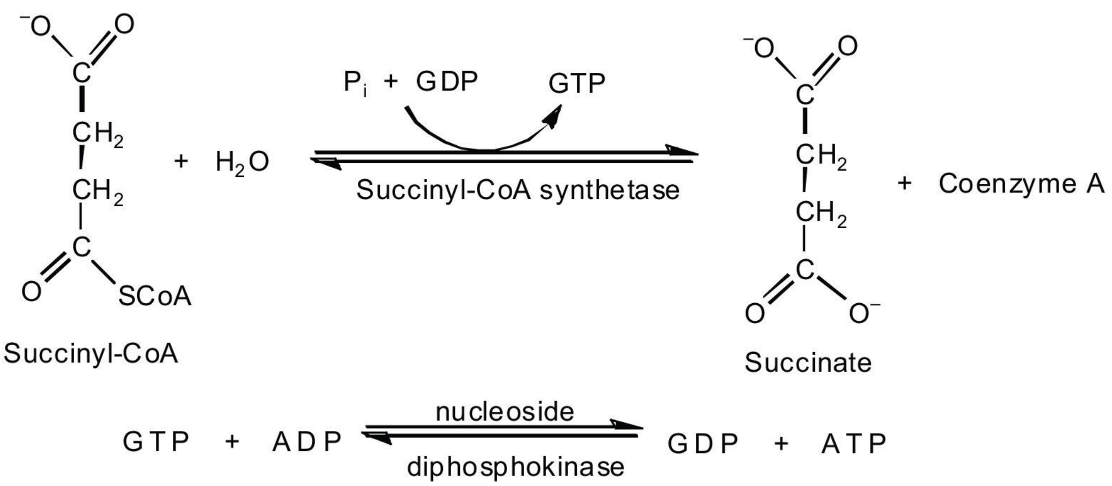
NOTE **α-KGDH complex contains five Coenzymes that act as carriers or oxidants for the intermediates of the reactions. E1 requires TPP, E2 requires lipoic acid and CoA and E3 requires NAD+ and FAD.**

Mechanism of action

Initially α-ketoglutarate and thiamine pyrophosphate (TPP or vitamin B_1) are bound by α-ketoglutarate acid dehydrogenase subunits, then α-ketoglutarate is decarboxylated by α-ketoglutarate acid dehydrogenase with help from TPP.
E_2 (transacetylase with cofactor lipoate) transfers substrate to CoA, forming succinyl CoA. Lipoate has 2 sulfur atoms connected by a disulfide bond, E_1 donates the decarboxylated α-KG to one of the sulfur atoms causing the breaking of the disulfide bridge. The other sulfur atom converts to thiol group by combining with H+ from the solution. (succinate combines with CoA because when a carboxylic group is being released as a CO2 molecule the adjacent carbon becomes terminal, thus succinate becomes reactive and combines with anything to hold it and the best acyl carrier molecule is CoenzymeA. Then E2 (transacetylase) becomes oxidized and the electrons are being loaded to FAD molecule producing FADH2.

Step 5

the energy released by the hydrolysis of the high-energy thioester bond of succinyl-CoA is used to form guanosine triphosphate (GTP) from guanosine diphosphate (GDP) and inorganic phosphate in a reaction catalyzed by *succinate thiokinase* in a process called substrate-level phosphorylation. This step is the only reaction in the citric acid cycle that directly forms a high-energy phosphate compound. GTP can readily transfer its terminal phosphate group to adenosine diphosphate (ADP) to generate ATP. Most of the ATP produced by respiration results later, from oxidative phosphorylation, when the NADH and FADH2 produced relay the electrons to the ETC.



Step 6

Succinate dehydrogenase then catalyzes the removal of two hydrogen atoms from succinate, forming fumarate (ALKENE). This oxidation-reduction reaction uses flavin adenine dinucleotide (FAD), rather than NAD^+, as the oxidizing agent.

Step 7

In this step, a molecule of water is added to the double bond of fumarate to form malate (2° ALCOHOL) in a reaction catalyzed by *fumarase.*

Step 8

One revolution of the cycle is completed with the oxidation of Alcohol group of malate to a keto group forming oxaloacetate, brought about by *malate dehydrogenase*. This is the third oxidation-reduction reaction that uses NAD^+ as the oxidizing agent.
Oxaloacetate can accept an acetyl group from acetyl-CoA, allowing the cycle to begin again.

ELECTRON carrying molecules	
NAD^+	FAD
Pair of electrons (**H**$^-$), same source	Single electrons (H•), different sources
Alcohols to ketones by malate dehydrogenase & isocitrate dehydrogenase	Succinate to fumarate, lipoate to lipoate disulfide in α-KGDH
NADH plays a regulatory role in balancing energy metabolism	FAD must remain tightly, sometimes covalently, attached to its enzyme
	***E*°** for enzyme-bound FAD varies

YOU GOTTA SOLVE THIS

1) If the Krebs cycle is overstimulated, the body will produce too much of which of the following molecules?
A) Oxygen
B) Glucose
C) Carbon dioxide
D) Acetyl CoA
E) Pyruvate

2) MO'TASEM ABU JABER took a neural sample and separated the cell body from the axon. He noticed that when he placed both parts on a pyruvate plate, the cell body began releasing carbon dioxide. What could explain the result?
A) The cell body contains mitochondria
B) The carbon dioxide is used as a messenger to communicate with the axon
C) The cell body is degrading
D) The carbon dioxide came from the plate
E) None of these

3) In the citric acid cycle, a flavin coenzyme is required for?
A) condensation of acetyl-CoA and oxaloacetate
B) oxidation of fumarate
C) oxidation of isocitrate
D) oxidation of malate
E) oxidation of succinate

4) All of the oxidative steps of the citric acid cycle are linked to the reduction of NAD+ except:

A) isocitrate dehydrogenase
B) α-KG dehydrogenase
C) pyruvate dehydrogenase
D) succinate dehydrogenase

5) For the following reaction,

Maltate + $NAD^+ \longrightarrow$ Oxaloacetate + NADH , $\Delta G°= +29.7$ kj/mol.

The reaction:

A) can never occur in a cell
B) can only occur in a cell if it is coupled to another reaction for which ΔG° is positive
C) can only occur in a cell in which NADH is converted to NAD+ by electron transport
D) may occur in cells at certain concentrations of substrate and product
E) would always proceed at a very slow rate

6) The reaction of the citric acid cycle that produces an ATP equivalent (in the form of GTP) by substrate-level phosphorylation is the conversion of?

A) citrate to isocitrate
B) fumarate to malate
C) malate to oxaloacetate
D) succinate to fumarate
E) succinyl-CoA to succinate

7) Which one of the following enzymatic activities would be decreased by thiamine deficiency?

A) Fumarase
B) Isocitrate dehydrogenase
C) Malate dehydrogenase
D) Succinate dehydrogenase
E) α-Ketoglutarate dehydrogenase complex

8) The two moles of CO2 produced in the first turn of the citric acid cycle have their origin in the?

A) carboxyl and methylene carbons of oxaloacetate
B) carboxyl group of acetate and a carboxyl group of oxaloacetate
C) carboxyl group of acetate and the keto group of oxaloacetate
D) two carbon atoms of acetate
E) two carboxyl groups derived from oxaloacetate

9) Oxaloacetate uniformly labeled with ^{14}C (i.e., with equal amounts of ^{14}C in each of its carbon atoms) is condensed with unlabeled acetyl-CoA. After a single pass through the citric acid cycle back to oxaloacetate, what fraction of the original radioactivity will be found in the oxaloacetate:

A) 1/4
B) 1/2
C) 3/4
D) all
E) 0

10) Malonate is a competitive inhibitor of succinate dehydrogenase. If malonate is added to a mitochondrial preparation that is oxidizing pyruvate as a substrate, which of the following compounds would you expect to decrease in concentration:

A) Citrate
B) Fumarate
C) Isocitrate
D) Pyruvate
E) Succinate

11) In mammals, each of the following occurs during the citric acid cycle except:

A) formation of α-ketoglutarate
B) generation of NADH and FADH
C) metabolism of acetate to carbon dioxide and water
D) net synthesis of oxaloacetate from acetyl-CoA
E) oxidation of acetyl-CoA

12) Acetyl-CoA labeled with ^{14}C in both of its acetate carbon atoms is incubated with unlabeled oxaloacetate and a crude tissue preparation capable of carrying out the reactions of the citric acid cycle. After one turn of the cycle, oxaloacetate would have ^{14}C in:

A) all four carbon atoms.
B) no pattern that is predictable from the information provided
C) none of its carbon atoms
D) the keto carbon and one of the carboxyl carbons
E) the two carboxyl carbons.