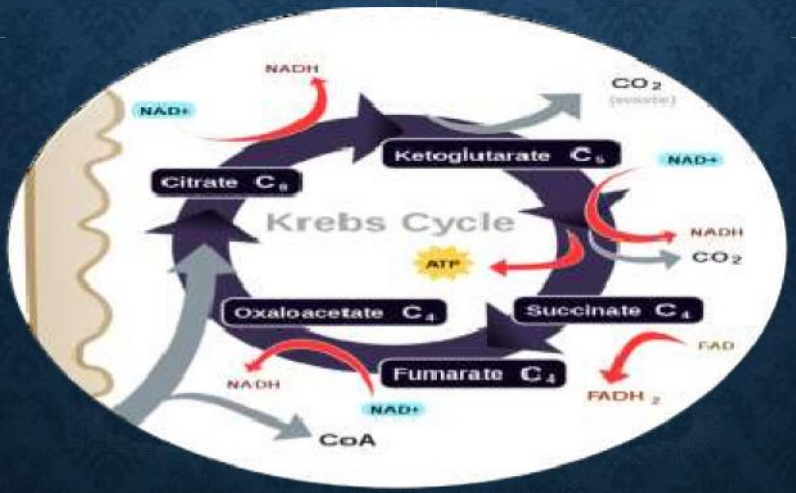
Biochemistry

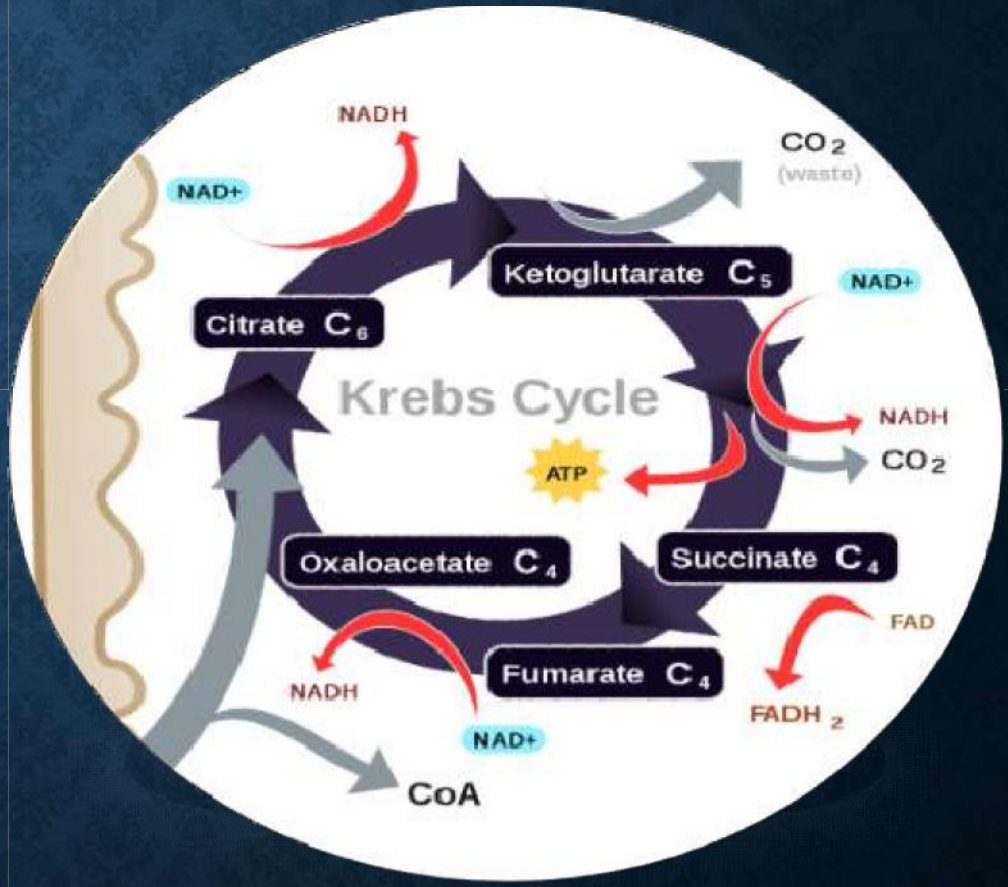
Citric acid cycle (TCA)



By:
Dr. Canan Can
Omeed akbar ali

CITRIC ACID CYCLE

- Introduction
- Site
- Pathway
- Energetics
- Regulation
- Clinical importance

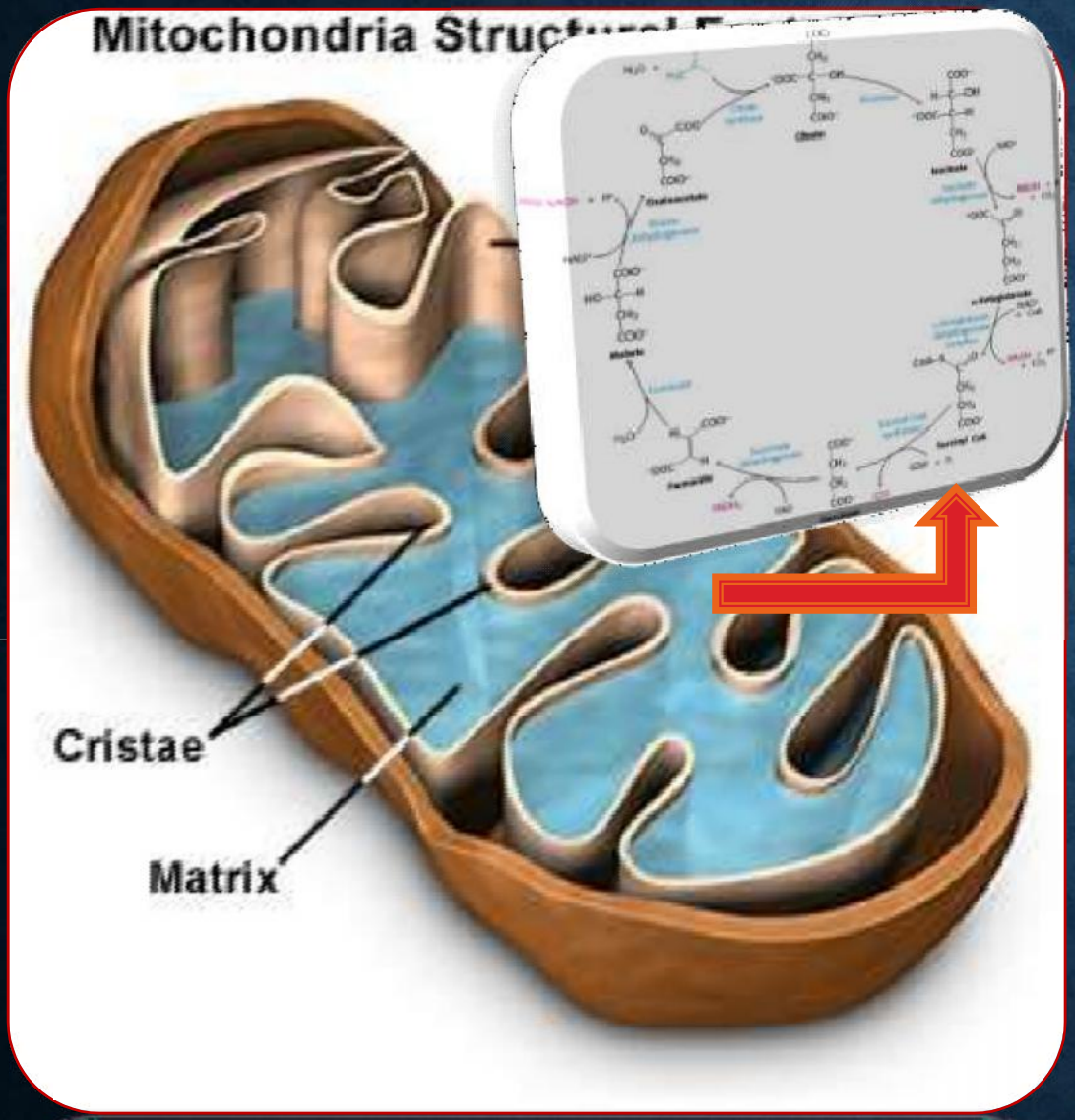


INTRODUCTION

- The citric acid cycle is the central metabolic hub of the cell.
- It is the final common pathway for the oxidation of fuel molecule such as amino acids, fatty acids, and carbohydrates.
- In eukaryotes, the reactions of the citric acid cycle take place inside mitochondria, in contrast with those of glycolysis, which take place in the cytosol.

Definition :

- The citric acid cycle is a series of reactions that brings about catabolism of acetyl-coA liberating **reducing equivalents** which upon oxidation through respiratory chain of mitochondria, **generate ATP.**
- It plays a central role in the breakdown or **catabolism** of organic fuel molecules—i.e **glucose** and some other sugars, fatty acids, and some amino acids. Before these rather large molecules can enter the TCA cycle they must be degraded into a two-carbon compound called acetyl coenzyme A (acetyl CoA). Once fed into the TCA cycle, acetyl CoA is converted into **carbon dioxide** and energy.

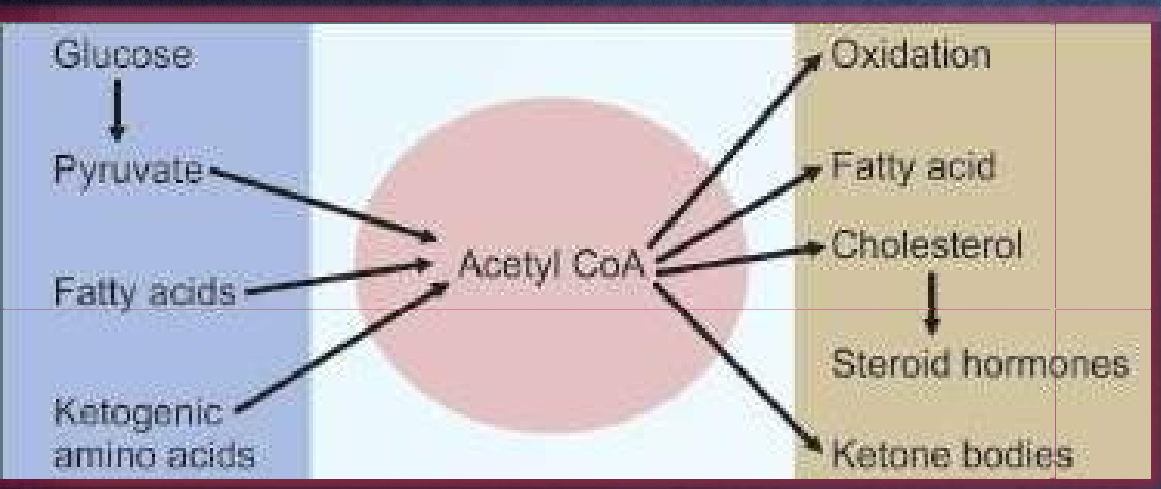


- Takes place in the matrix of the mitochondria.
- It happens once for every pyruvate molecule in glycolysis….

❑ Purpose

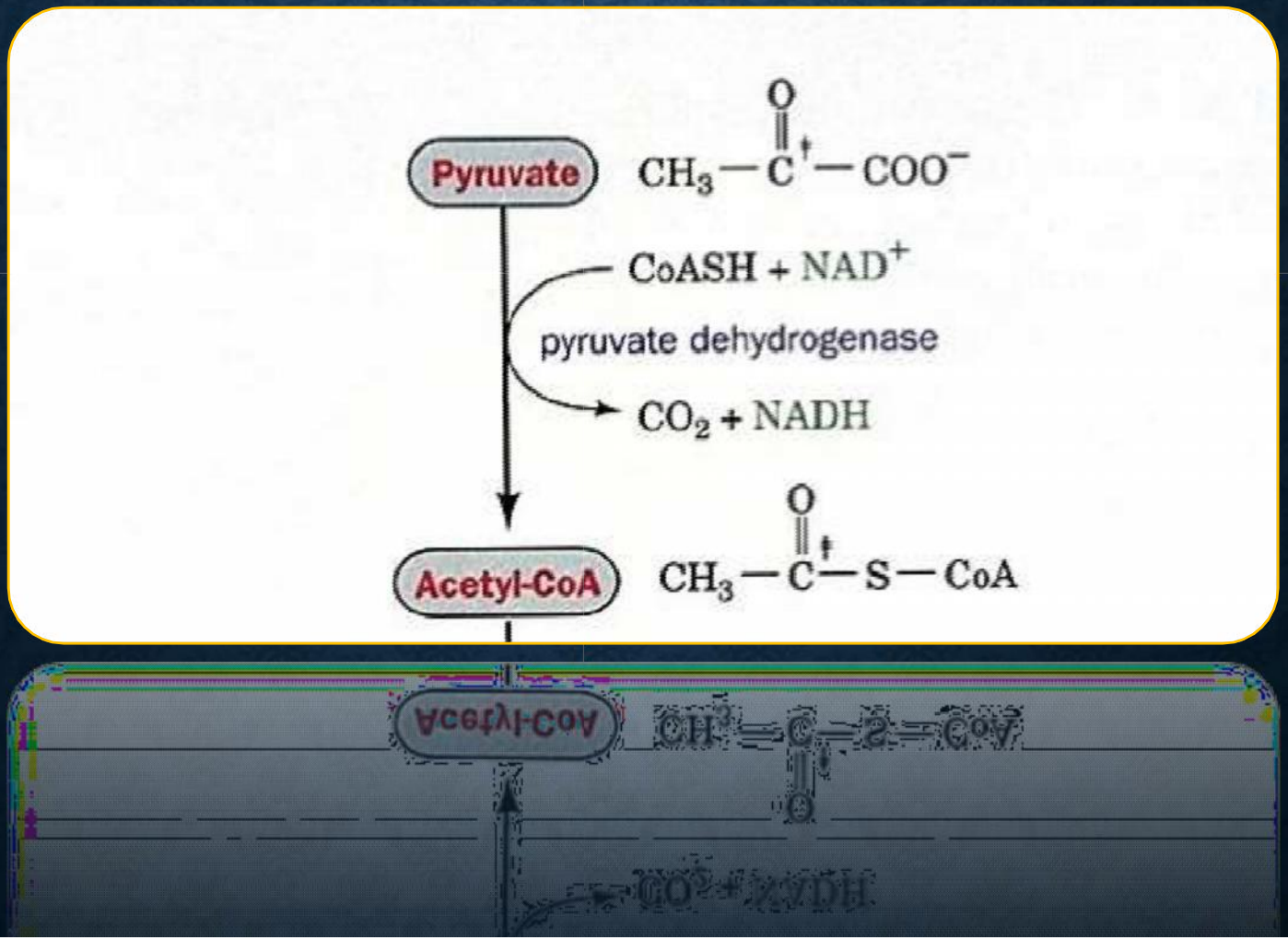
- Conversion of Acetyl-CoA to CO_2
- Generates reducing equivalents (NADH + H^+, FADH2) & GTP to be oxidized in the respiratory chain to generate ATP

OVERVIEW

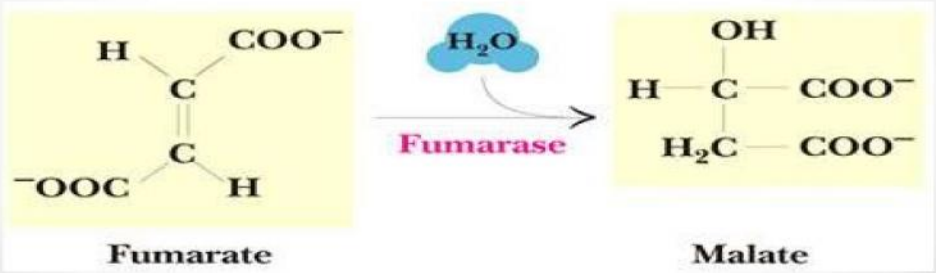


- **Acetyl coA, the precursor for fatty acid synthesis is produced from pyruvate, ketogenic amino acids, fatty acid oxidation and by alcohol metabolism.**
- **It is a substrate for TCA cycle and a precursor for faty acids ketone bodies and sterols.**

STEPS INVOLVED IN TCA CYCLE



Add water to Fumarase



Fumarate

Malate

COO

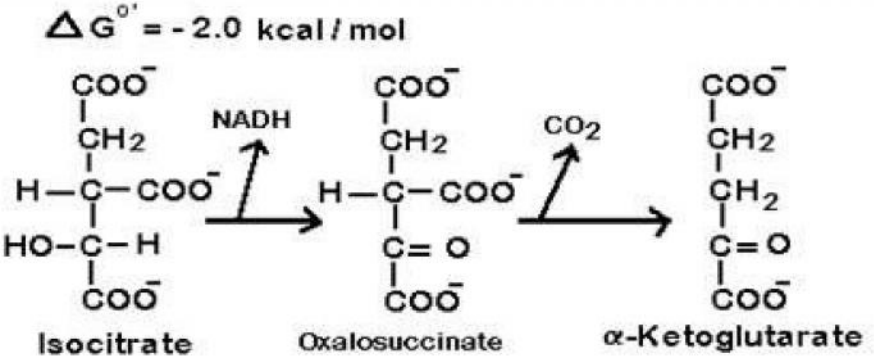
α-Ketoglutarate

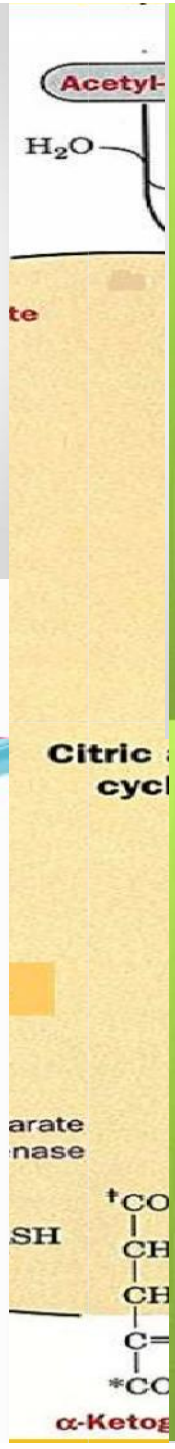
S—CoA

Succinyl CoA

Isocitrate Dehydrogenase

Oxidation and the removal the carboxyl of Citrate.



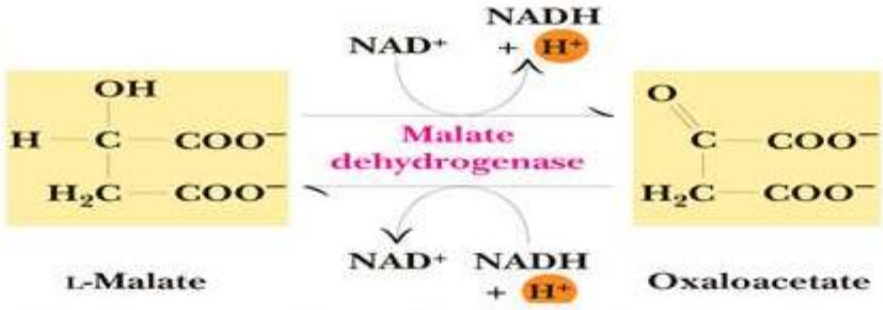


Step:10 Conversion of malate to oxaloacetate

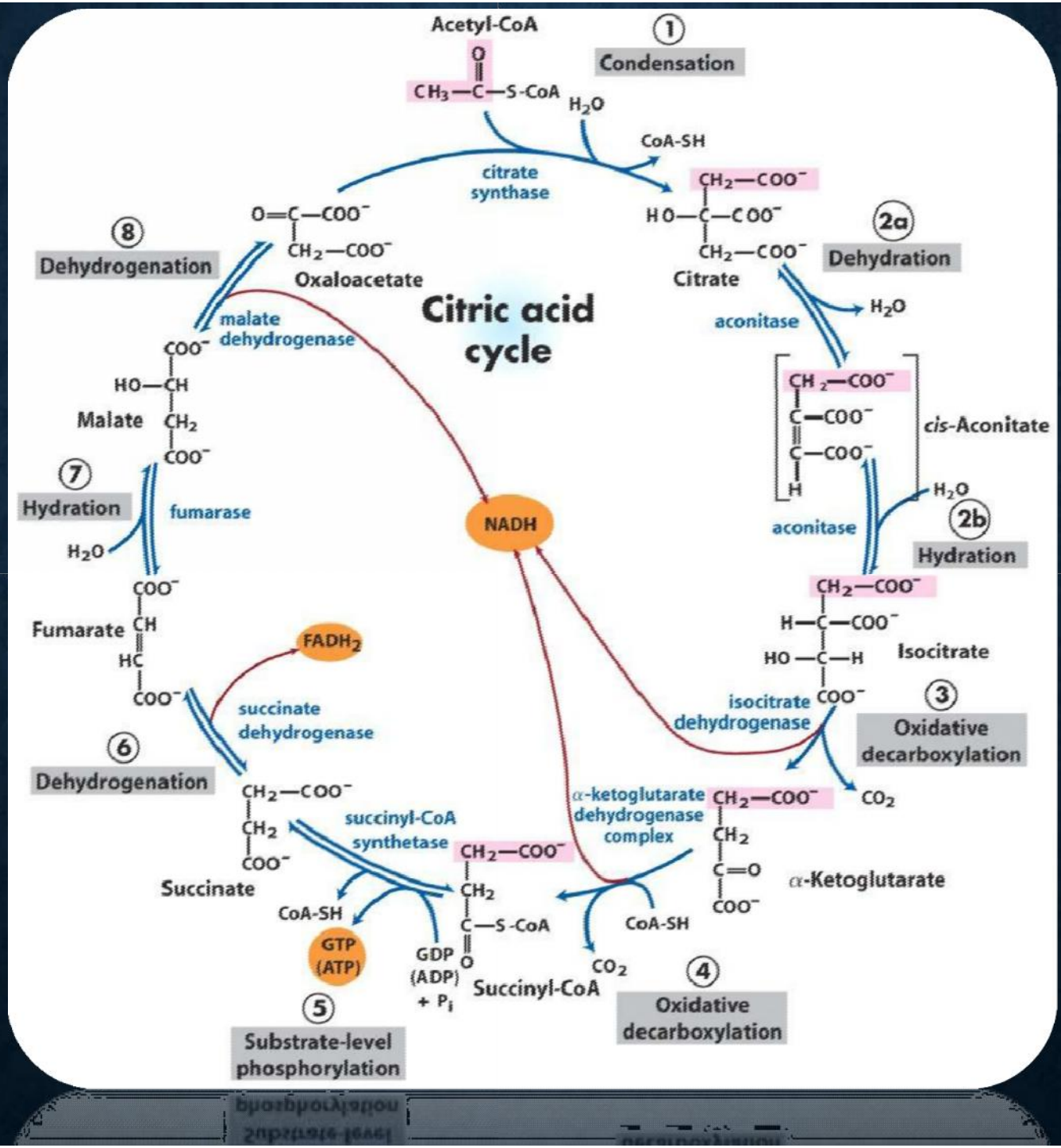
- Malate is then oxidized to oxaloacetate by malate dehydrogenase.
- The third & final synthesis of NADH occurs at this stage.
- The oxaloacetate is regenerated which can combine with another molecule of acetyl CoA & continue the cycle.

Malate Dehydrogenase

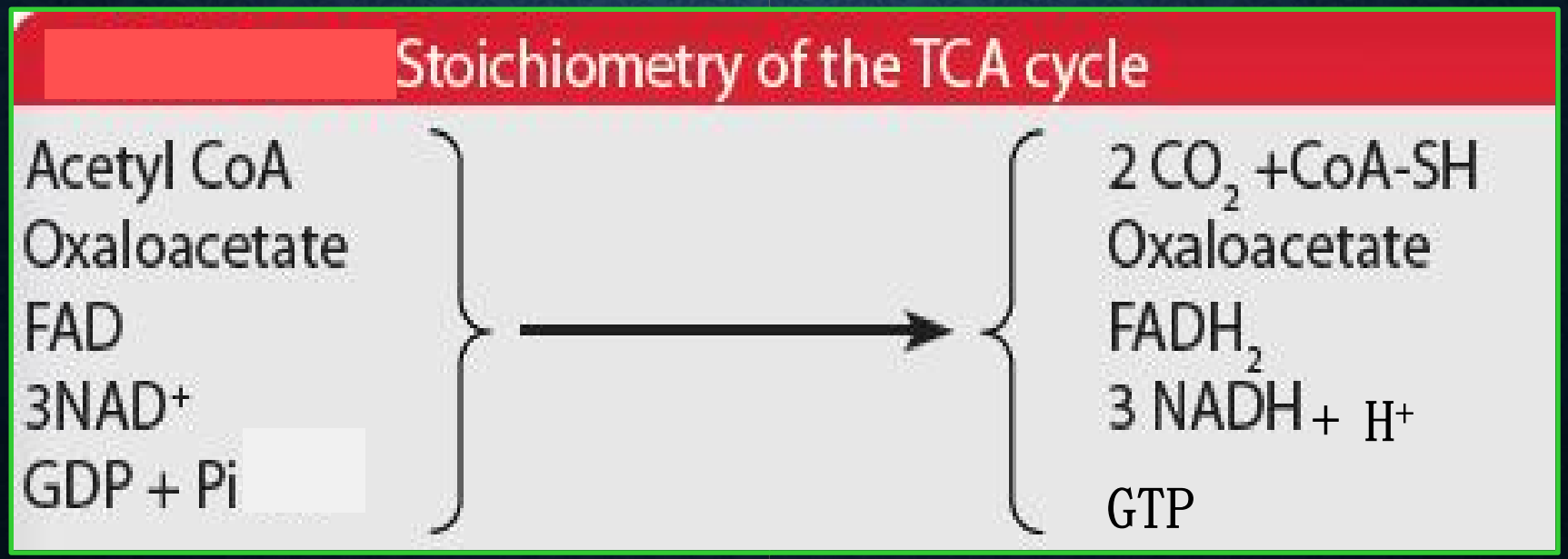
Oxidation of malate



–This reaction generates a three of the partial NADH cycle


Citric acid cycle
Acetyl-CoA
1
Condensation
citrate synthase
CoA-SH
Oxaloacetate
Citrate
2a
Dehydration
aconitase
cis-Aconitate
2b
Hydration
Isocitrate
isocitrate dehydrogenase
3
Oxidative decarboxylation
α-ketoglutarate dehydrogenase complex
α-Ketoglutarate
4
Oxidative decarboxylation
Succinyl-CoA
succinyl-CoA synthetase
5
Substrate-level phosphorylation
Succinate
succinate dehydrogenase
6
Dehydrogenation
Fumarate
7
Hydration
fumarase
Malate
8
Dehydrogenation
malate dehydrogenase
NADH
FADH2
GTP (ATP)
GDP (ADP) + Pi
CO2
H2O

- TCA cycle is an open cycle
- Operates only under aerobic conditions
- This is the Final common pathway of oxidative metabolism
- Two carbon dioxide molecules are released as a waste product of respiration



Energetics : 2 Acetyl CoA from 2 Pyruvate

Acetyl-CoA + 3 NAD^+ + [FAD] + GDP + Pi + 2 H_2O ⟷ CoASH + 3 NADH + 3 H^+ + [$FADH_2$] + GTP + 2 CO_2

- 1NADH+H^+ = 3/2.5 ATP
- 1$FADH_2$ = 2/1.5 ATP
- 1GTP = 1 ATP

3. Number of ATP generated from GTP	1
	12
2. Number of ATP generated by oxidation of $FADH_2$	2
3. Number of ATP generated from GTP	1
	12

×2=24

ATP aeneration steps

Step No	*Reactions*	*Co-enzyme*	*ATPs (old-calculation)*	*ATPs (new calculation)*
3	Isocitrate → alpha keto glutarate	NADH	3	2.5
4	Alpha keto glutarate → succinyl CoA	NADH	3	2.5
5	Succinyl CoA→Succinate	GTP	1	1
6	Succinate → Fumarate	$FADH_2$	2	1.5
8	Malate → Oxalo acetate	NADH	3	2.5
		Total	12	10

ATP generation during oxidation of Glucose

Process	Number of ATP/mol of glucose
1. Glycolysis	8 / 7
2. Pyruvate dehydrogenase	6 / 5
3. Citric acid cycle	24 / 20
Total	38 / 32

- ✓Net ATP production depends on shuttle used for the transfer of reducing equivalents from cytosol to mitochondria.

SIGNIFICANCE OF TCA CYCLE:

1. Complete oxidation of Acetyl CoA
2. As provider of energy
3. Final common oxidative pathway
4. Integration of major metabolic pathways
5. Fat is burned on the wick of carbohydrates
6. Excess carbohydrates are converted to Neutral fat
7. No net synthesis of carbohydrates from fat
8. Amino acids enters TCA cycle
9. Amphibolic pathway
10. Anaplerotic role

Bio medical importance

- Bioenergetics is the study of the energy changes accompanying biochemical reactions. Biologic systems are essentially isothermic and use chemical energy to power living processes.
- Animal obtains suitable fuel from its food to provide the energy for metabolism.
- Death ⟹ starvation

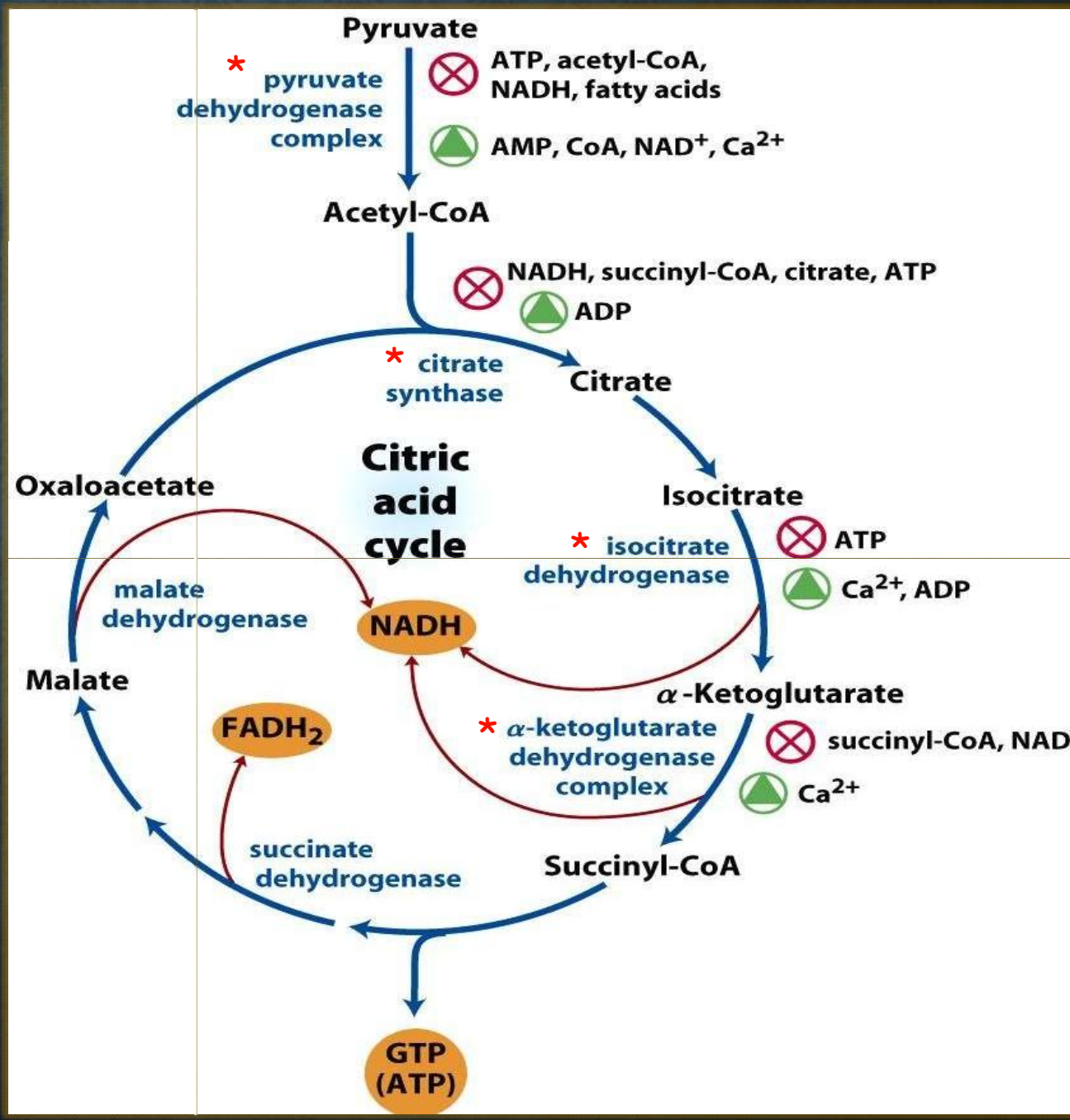
Bio medical importance

- **Thyroid hormones control the rate of energy release and disease results when they malfunction.**
- **Excess storage of surplus energy causes obesity.**

- This cycle is a series of chemical intermediates.
- Each step is catalyzed by a specific enzyme.

Regulation of TCA cycle

- Indicator molecules of higher energy state i.e. ATP, NADH, citrate, Acetyl CoA – inhibit TCA cycle
- Indicator molecules of low energy state i.e. ADP, AMP, NAD^+ – stimulate TCA cycle



Regulation of TCA cycle enzymes

a)Citrate synthase- There is allosteric inhibition of citrate synthase by ATP and long-chain fatty acyl-CoA.

b)Isocitrate dehydrogenase- is allosterically stimulated by ADP, which enhances the enzyme's affinity for substrates. In contrast, NADH inhibits iso-citrate dehydrogenase by directly displacing NAD+. ATP, too, is inhibitory.

Regulation of TCA cycle enzymes

c) α-ketoglutarate dehydrogenase -α- Ketoglutarate dehydrogenase is inhibited by succinyl CoA and NADH. In addition, α-ketoglutarate dehydrogenase is inhibited by a high energy charge. Thus, the rate of the cycle is reduced when the cell has a high level of ATP.

d)Succinate dehydrogenase is inhibited by oxaloacetate, and the availability of oxaloacetate, as controlled by malate dehydrogenase, depends on the [NADH]/[NAD+] ratio.

INHIBITORS OF TCA CYCLE

1. Fluoroacetate
   - Condensation FluoroacetylCoA with Oxaloacetate → Fluorocitrate → inhibit Aconitase enzyme → accumulation of citrate
   - Fluoroacetate → pesticide
2. Malonate → Succinate dehydrogenase enzyme
3. Arsenite → α-ketoglutarate dehydrogenase enzyme

METABOLIC DEFECTS

- Extremely rare

1. Defect in PDH

   - Lactic acidosis

   - Neurologycal dosorders

2. Defect In Pyruvate carboxylase

   - ↓ Oxaloacetate

   - Hyperammonemia

   - Lactic acidosis

   - Hyperalaninemia.

WHY TCA IS CALLED AMPHIBOLIC?

It plays both catabolic and anabolic role.

***Catabolic role**:

Acetyl CoA is oxidized to C02, H2O giving out energy.

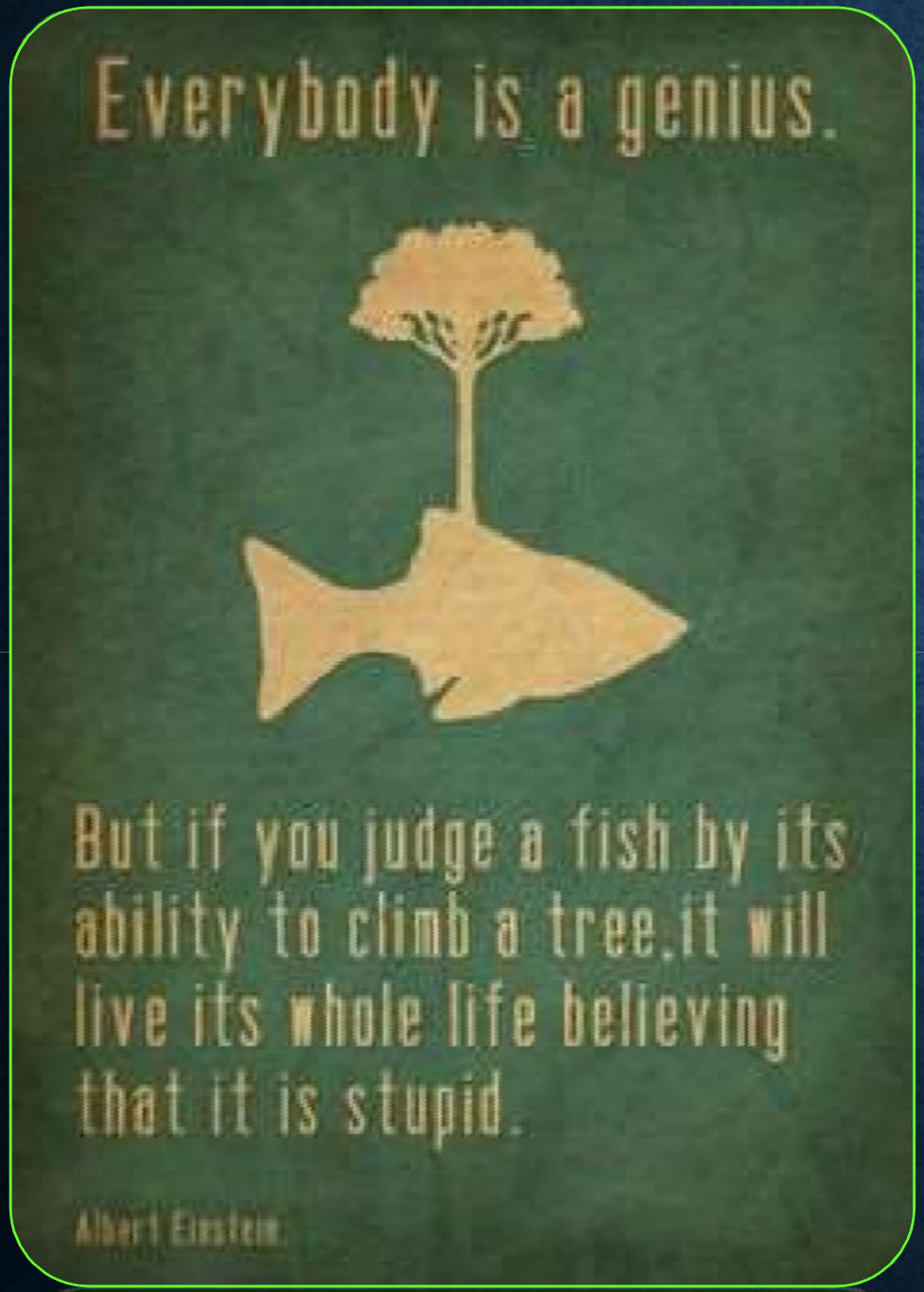
***Anabolic role**:

Intermediates of TCA cycle plays a role in synthesis like heme formation, FA synthesis, Cholesterol, Steroid synthesis.

SUMMARY

- Pyruvate is converted to acetyl-CoA by the action of p yruvate dehydrogenase complex, a huge enzyme complex.
- Acetyl-CoA is converted to 2 CO_2 via the eight-step citric acid cycle, generating three NADH, one $FADH_2$, and one ATP (by substrate-level phophorylation).

- Intermediates of citric acid cycle are also used as biosynthetic precursors for many other biomolecules, including fatty acids, steroids, amino acids, heme, pyrimidines, and glucose.
- Oxaloacetate can get replenished from pyruvate, via a carboxylation reaction catalyzed by the biotin-containing pyruvate carboxylase.


Everybody is a genius.
But if you judge a fish by its ability to climb a tree, it will live its whole life believing that it is stupid.
Albert Einstein

THANK YOU

Let’s have a break