From Last Lecture

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- Regenerating NAD^+ from pyruvate metabolism keeps glycolysis running by ensuring a steady supply of this essential electron carrier.

- Pyruvate is like a Swiss army knife—it can be converted into many things, depending on what the cell needs to survive or grow.

- Glycolysis is the gateway to understanding how energy flows through a cell
- Phosphorylating glucose adds a molecular 'anchor'—it keeps glucose inside the cell and marks it for metabolism

- Fructose and galactose are like alternate passengers—they take different entry ramps but eventually merge onto the glycolysis highway

In case you did not appreciate Glycolysis yet…

- **Central Role in Metabolism**
  - first step in glucose metabolism, occurring in nearly all living organisms.
  - connects to many other pathways: fermentation, the citric acid cycle, oxidative phosphorylation, and biosynthetic routes.
- **Showcases major biochemical principles**
  - enzyme catalysis
  - energy coupling (e.g., ATP generation and usage)
  - redox reactions (e.g., NAD^+ to NADH)
  - regulation of metabolism (e.g., feedback inhibition)
- **Biomedical Relevance**
  - aberrant glycolysis is involved in diseases like and diabetes.
  - many glycolytic enzymes are drug targets.
- **Evolutionary Perspective**
  - glycolysis is ancient and highly conserved, hinting at early life's metabolic strategies.
  - occurs in the cytoplasm and doesn't require oxygen—useful to discuss anaerobic vs. aerobic life.

Metabolism in Context: Glycolysis Helps Pancreatic Beta Cells Sense Glucose

- Insulin is secreted by β cells of the pancreas in response to high blood levels of glucose. This secretion is stimulated by the metabolism of glucose by the β cells.
- Glucose enters β cells through GLUT2 and is metabolized to pyruvate, and the pyruvate is subsequently oxidized to CO_2 and H_2O.
- The increase in ATP closes a K^+ channel, which alters the charge across the cell membrane. This alteration in turn opens Ca^{2+} channels. The influx of Ca^{2+} ions stimulates the release of insulin.

- Insulin release is controlled by blood glucose levels.
- Glucose metabolism(glycolysis) increases ATP production. Higher ATP closes an ATP-sensitive potassium channel.
- This changes the membrane charge, opening a calcium channel.
- Calcium influx triggers insulin granules to fuse with the membrane.
- Insulin is released into the blood.

Figure 16.19
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Is pyruvate fermentation to ethanol present in humans?

Why do some people develop insulin resistance?

How is glycolysis regulated?

The Glycolytic Pathway is Tightly Controlled

- Irreversible steps in glycolysis are major control points: hexokinase, PFK-1, pyruvate kinase
- These enzymes are regulated in three main ways:
  - binding of allosteric effectors (milliseconds) or covalent modification (seconds)
  - transcriptional control (hours)

- In muscles, signal is AMP/ATP ratio

Phosphofructokinase

- The binding of ATP lowers the enzyme's affinity for fructose 6-phosphate.
- AMP reverses the inhibitory action of ATP.
- AMP competes with ATP for the binding site but does not inhibit the enzyme when bound.
- As a result, enzyme activity increases when the ATP/AMP ratio is lowered.

Phosphofructokinase from *E. coli*.

If glycolysis breaks down glucose to make energy, why doesn't the cell just reverse the same steps to make glucose when needed?

Which of the following statements best explains why glycolysis cannot simply run in reverse?

A. The enzymes used in glycolysis are not present in liver cells
B. Glycolysis includes irreversible steps that need alternative pathways
C. Glycolysis requires oxygen, but gluconeogenesis does not
D. Glucose is only made in the mitochondria

Answer: B

During fasting, the liver performs gluconeogenesis to:
A. Store excess energy
B. Convert glucose into glycogen
C. Maintain blood glucose levels for glucose-dependent tissues
D. Break down fats into glucose

Answer: C

Gluconeogenesis – The Basics

- Gluconeogenesis synthesizes glucose from noncarbohydrate precursors—an essential "recycling" process.
- Vital during fasting to supply glucose to the brain and red blood cells.
- Not simply the reverse of glycolysis
  - Irreversible glycolysis steps are bypassed with alternative exergonic reactions.
- Glycolysis and gluconeogenesis are reciprocally regulated to prevent energy-wasting futile cycles.

- Also recycles lactate produced by active tissues.
- Primary site: liver; minor sites: kidney and other tissues.
- Not active in: brain, skeletal muscle, or heart—these rely on blood glucose.

The Gluconeogenic Pathway

- Gluconeogenesis synthesizes glucose from noncarbohydrate precursors.

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The Gluconeogenic Pathway

The Gluconeogenic Pathway

Gluconeogenesis is NOT the reversal of Glycolysis

$$\text{Glucose} + \text{ATP} \xrightarrow{\text{Hexokinase}} \text{glucose 6-phosphate} + \text{ADP}$$

$$\Delta G = -33\ \text{kJ mol}^{-1}\left(-8.0\ \text{kcal mol}^{-1}\right)$$

$$\text{Fructose 6-phosphate} + \text{ATP} \xrightarrow{\text{Phosphofructokinase}} \text{fructose 1,6-bisphosphate} + \text{ADP}$$

$$\Delta G = -22\ \text{kJ mol}^{-1}\left(-5.3\ \text{kcal mol}^{-1}\right)$$

$$\text{Phosphoenolpyruvate} + \text{ADP} \xrightarrow{\text{Pyruvate kinase}} \text{pyruvate} + \text{ATP}$$

$$\Delta G = -17\ \text{kJ mol}^{-1}\left(-4.0\ \text{kcal mol}^{-1}\right)$$

Three irreversible steps in glycolysis that must be bypassed in gluconeogenesis.

First Step in Gluconeogenesis

$$HCO_3^- + ATP \leftrightharpoons HOCO_2\text{-}PO_3^{2-} + ADP$$

$$\text{Biotin-enzyme} + HOCO_2\text{-}PO_3^{2-} \rightarrow CO_2\text{-biotin-enzyme} + P_i$$

$$CO_2\text{-biotin-enzyme} + \text{pyruvate} \leftrightharpoons \text{biotin-enzyme} + \text{oxaloacetate}$$

- in mitochondria, catalyzed by pyruvate carboxylase (PC), which…
- requires biotin (vitamin B7) as a prosthetic group.
- Biotin carries activated CO_2 in carboxylation reactions.
- Note that acetyl-CoA must be bound to the enzyme for biotin carboxylation.

Carboxybiotin covalently bound to ε-amino group of a lysine

First Step in Gluconeogenesis

Charles Bruglag/FeaturePics.

- Biotin carboxylase forms carboxyphosphate and attaches CO_2 to biotin.
- Biotin carboxyl carrier protein carries covalently bound biotin.
  - Swings ~75 Å to reach the pyruvate carboxylase domain.
- Pyruvate carboxylase domain transfers CO_2 to pyruvate → oxaloacetate.
- Tetramerization domain helps subunits assemble.

Oxaloacetate ➜ Phosphoenolpyruvate

- Oxaloacetate must move to the cytoplasm
  - Is reduced to malate by mitochondrial malate dehydrogenase
- Malate crosses the mitochondrial membrane, then is reoxidized to back to oxaloacetate in the cytoplasm (by cytoplasmic NAD^+-linked malate dehydrogenase).

Note: This is both a decarboxylation and a phosphorylation reaction catalyzed by PEPCK.

Lactate
Some amino acids
Pyruvate
Pyruvate carboxylase
ATP, HCO_3
ADP + P_i
Some amino acids
Oxaloacetate
Phosphoenolpyruvate carboxykinase
GTP
GDP, CO_2
Phosphoenolpyruvate
Enolase
H_2O
2 x
2-Phosphoglycerate
Phosphoglycerate mutase
3-Phosphoglycerate
Phosphoglycerate kinase
ATP
ADP
1,3-Bisphosphoglycerate
Glycerol
CH_2OH
O=C
$CH_2OPO_3^{2-}$
Dihydroxyacetone phosphate
Glyceraldehyde 3-phosphate dehydrogenase
P_i , NAD^+
Triose phosphate isomerase
Glyceraldehyde 3-phosphate
Aldolase

Fructose 1,6-bisphosphate
Fructose 1,6-bisphosphatase
H_2O
P_i
Fructose 6-phosphate
Phosphoglucose isomerase
Glucose 6-phosphate
Glucose 6-phosphatase
H_2O
P_i
Glucose

Fructose 1,6-bisphosphate ➜ Fructose 6-phosphate

- Phosphoenolpyruvate is metabolized by the enzymes of glycolysis in the reverse direction until the next irreversible step, the hydrolysis of fructose 1,6-bisphosphate.

- FBPase is an allosteric enzyme ➜ primary regulation point of gluconeogenesis.
- Fructose 6-phosphate produced ➜ Glucose 6-phosphate

Generation of free glucose is an important control point

- The generation of free glucose, which occurs essentially only in the liver, is the final step in gluconeogenesis.
- Glucose 6-phosphate is transported into the lumen of the endoplasmic reticulum.
- Glucose 6-phosphatase, an integral membrane on the inner surface of the endoplasmic reticulum, catalyzes the formation of glucose from glucose 6-phosphate.
- Transporters transport Pi (T2) and glucose (T3) back to the cytoplasm.

Let's do some accounting….

Gluconeogenesis:

$$2\ \text{Pyruvate} + 4\ \text{ATP} + 2\ \text{GTP} + 2\ \text{NADH} + 2\ \text{H}^+ + 6\ \text{H}_2\text{O} \rightarrow$$

$$\text{glucose} + 4\ \text{ADP} + 2\ \text{GDP} + 6\ \text{P}_\text{i} + 2\ \text{NAD}^+$$

$$\Delta G^{\circ\prime} = -48\ \text{kJ mol}^{-1} (-11\ \text{kcal mol}^{-1})$$

Reversal of Glycolysis:

$$2\ \text{Pyruvate} + 2\ \text{ATP} + 2\ \text{NADH} + 2\ \text{H}^+ + 2\ \text{H}_2\text{O} \rightarrow$$

$$\text{glucose} + 2\ \text{ADP} + 2\ \text{P}_\text{i} + 2\ \text{NAD}^+$$

$$\Delta G^{\circ\prime} = +90\ \text{kJ mol}^{-1} (+22\ \text{kcal mol}^{-1})$$

Quick Quiz 2

Which of the following is true for both glycolysis and gluconeogenesis?

A. Some steps in the pathway consume ATP.
B. Some steps in the pathway produce ATP.
C. Glucose-6-phosphate is an intermediate in the pathway.
D. All of the above.
E. A & C

Gluconeogenesis and glycolysis in the liver are under reciprocal regulation

Reciprocal regulation → glycolysis will predominate when glucose is abundant, and gluconeogenesis takes over when glucose is scarce.

A signal molecule that is a potent activator of phosphofructokinase.

Gluconeogenesis and glycolysis in the liver are under reciprocal regulation

- The balance between glycolysis and glycogenesis in the liver is sensitive to blood-glucose concentration.
- In liver, the rates of glycolysis and gluconeogenesis are adjusted to maintain blood glucose levels.
- The key regulator of glucose metabolism in liver is fructose 2,6-bisphosphate. Fructose 2,6-bisphosphate stimulates phosphofructokinase and inhibits fructose 1,6-bisphosphatase.

- The kinase that synthesizes fructose 2,6-bisphosphate and the phosphatase that hydrolyzes this molecule are located on the same polypeptide chain. Such an arrangement is called a bifunctional enzyme.

Gluconeogenesis and glycolysis in the liver are under reciprocal regulation

- Rates of glycolysis and gluconeogenesis in the liver are sensitive to blood-glucose concentration and are adjusted to maintain blood glucose levels.
- The key regulator of glucose metabolism in liver is fructose 2,6-bisphosphate. Fructose 2,6-bisphosphate stimulates phosphofructokinase and inhibits fructose 1,6-bisphosphatase.

Gluconeogenesis and glycolysis are reciprocally regulated

- Insulin normally inhibits gluconeogenesis. In type 2 diabetes, insulin fails to act, a condition called insulin resistance.
- The enzymes of gluconeogenesis, especially PEPCK (phosphoenolpyruvate carboxykinase), are active leading to abnormally high levels of blood glucose.
- Exercise and diet can enhance insulin sensitivity

Clinical Insight

Insulin Fails to Inhibit Gluconeogenesis in Type 2 Diabetes

http://www.medicinenet.com/type_2_diabetes_pictures_slideshow/article.htm

Why do some people develop insulin resistance?

- Insulin resistance → cells no longer respond well to insulin, so glucose uptake is impaired, even though insulin is present.

- Insulin binds to its receptor
  - normally activates a signaling cascade to insert GLUT4 transporters on the membrane.
- In insulin resistance
  - the signaling pathway is disrupted.
  - GLUT4 transporters don't reach the surface, so glucose can't enter the cell efficiently.

- Causes: fat buildup in liver and muscle cells, genetics, chronic high insulin levels, inflammation and oxidative stress

- As a result, blood glucose stays high, pancreas works harder → insulin increases; over time → Beta-cell fatigue → Type 2 Diabetes

Metabolism in context: Precursors Formed by Muscle Are Used by Other Organs

- Muscle and liver display inter-organ cooperation in a series of reactions called the ***Cori cycle***.
- Lactate produced by muscle during contraction is released into the blood.
- Liver removes the lactate and converts it into glucose, which can be released into the blood.

Quick Quiz 3

What is the name of the enzyme that catalyzes the conversion of pyruvate to lactate in muscle cells?

A. pyruvate decarboxylase
B. lactate decarboxylase
C. pyruvate dehydrogenase
D. lactate dehydrogenase
E. None of the above.

Quick Quiz 4

Which of the following occurs during glycolysis?

A. A six-carbon molecule is split into two different three-carbon molecules.
B. A three-carbon molecule combines with another three-carbon molecule to form a six-carbon molecule.
C. NAD^+ participates in an oxidation reaction.
D. A & C
E. B & C

Answer:
D

Quick Quiz 1

Glucose is an essential fuel for the brain. In times of starvation, glucose is generated from non-diet precursors. As skeletal muscle breaks down, _____ is/are generated and enter the gluconeogenic pathway to generate fuel for the brain.

A. nitrogen
B. lactate
C. urea
D. amino acids
E. glycerol

From Glycolysis to Gluconeogenesis

Glycolysis – Energy Generation

➜ breakdown of glucose to pyruvate
➜ occurs in the cytoplasm
➜ produces ATP and NADH
➜ active during fed state
➜ Key enzymes:
- Hexokinase
- Phosphofructokinase-1 (PFK-1)
- Pyruvate kinase

While glycolysis breaks down glucose to meet immediate energy demands, gluconeogenesis allows the body to maintain glucose levels when dietary intake is low

Gluconeogenesis – Glucose Synthesis

➜ synthesis of glucose from non-carbohydrate precursors
➜ occurs mainly in liver (and kidney)
➜ consumes ATP and NADH
➜ active during fasting or prolonged exercise
➜ bypasses glycolysis's irreversible steps using:
- Pyruvate carboxylase
- PEP carboxykinase
- Fructose 1,6-bisphosphatase
- Glucose 6-phosphatase

Assigned Problems

Chapter	Tymochko, Berg, Stryer, Biochemistry, 2nd Edition,	**Chapter**	Tymochko, Berg, Stryer, Biochemistry, 2nd Edition,
16	1, 3, 9, 10, 15, 17, 19, 22, 24, 25, 29, 31.	17	2, 4, 5, 8, 10, 11, 17, 20, 21, 22.
Chapter	Tymochko, Berg, Stryer, Biochemistry, 3rd Edition, 4th Edition (bottom line)	**Chapter**	Tymochko, Berg, Stryer, Biochemistry, 3rd Edition, 4th Edition (bottom line)
16	2, 4, 10, 11, 15, 17, 19, 22, 24, 26, 31, 33.	17	2, 4, 5, 8, 10, 11, 17, 22, 23, 24.

Lecture 10

The Citric Acid Cycle

https://www.youtube.com/watch?v=JPCs5pn7UNI

The Organic Mechanisms of the Coenzymes

Lecture Outline:

- Pyruvate Dehydrogenase Forms Acetyl Coenzyme A from Pyruvate
- The Pyruvate Dehydrogenase Complex Is Regulated by Two Mechanisms
- The Citric Acid Cycle Consists of Two Stages
- The Citric Acid Cycle: Stage One
- The Citric Acid Cycle: Stage Two
- The Citric Acid Cycle Is Regulated
- The Glyoxylate Cycle Enables Plants and Bacteria to Convert Fats into Carbohydrates

Readings:

Tymochko, Berg, Stryer,
Biochemistry, 2nd Edition,
Ch. 18, 19, pp. 317 – 346
3rd Ed., Ch. 18,19, pp. 329 - 354
4th Ed., Ch. 18,19, pp. 363 - 387

- Pyruvate is like a Swiss army knife–it can be converted into many things, depending on what the cell needs to survive or grow.

Cellular Respiration

Cellular Respiration

https://allinonehighschool.com/aerobic-cellular-respiration/

One-way traffic facilitates traffic flow and directs flow toward specific locations. Similarly, pyruvate dehydrogenase directs pyruvate into the formation of acetyl CoA: it is the one-way link between glycolysis and cellular respiration.

The Krebs Cycle/The Citric Acid Cycle (CAC)/ The Tricarboxylic Acid Cycle (TAC)

Phosphoryl-transfer potential is an important form of cellular energy transformation

Compounds with high phosphoryl-transfer potential can be used to make ATP from ADP.

The Krebs Cycle/The Citric Acid Cycle (CAC)/ The Tricarboxylic Acid Cycle (TAC)

The Krebs Cycle/The Citric Acid Cycle (CAC)/ The Tricarboxylic Acid Cycle (TAC)

Preparation for the Krebs Cycle

- Under aerobic conditions, pyruvate enters the mitochondria where it is converted into ***acetyl CoA***.
- Acetyl CoA is the fuel for the citric acid cycle, which processes the two-carbon acetyl unit to 2 molecules of CO_2 while generating high-energy electrons that can be used to form ATP.

This thioester linkage is a "high energy" bond, which is particularly reactive. Hydrolysis of the thioester bond is exergonic(−31.5 kJ/mol)

Acetyl coenzyme A (Acetyl CoA)

Pyruvate Dehydrogenase Forms Acetyl Coenzyme A from Pyruvate

- The pyruvate dehydrogenase complex, a mitochondrial matrix enzyme, oxidatively decarboxylates pyruvate to form acetyl CoA.

$$\text{Pyruvate} + \text{CoA} + \text{NAD}^+ \rightarrow \text{acetyl CoA} + \text{CO}_2 + \text{NADH} + \text{H}^+$$

(Left) Omikron/Science Source.

- This reaction is an irreversible link between glycolysis and the citric acid cycle.

Pyruvate Dehydrogenase Forms Acetyl Coenzyme A from Pyruvate

The synthesis of acetyl coenzyme A from pyruvate requires three enzymes and five coenzymes.

- The synthesis of acetyl CoA from pyruvate consists of three steps: a decarboxylation, an oxidation, and the transfer to CoA.

Enzyme	Abbreviation	Prosthetic group	Reaction catalyzed
Pyruvate dehydrogenase component	E_1	TPP	Oxidative decarboxylation of pyruvate
Dihydrolipoyl transacetylase	E_2	Lipoamide	Transfer of acetyl group to CoA
Dihydrolipoyl dehydrogenase	E_3	FAD	Regeneration of the oxidized form of lipoamide

Pyruvate Dehydrogenase Forms Acetyl Coenzyme A from Pyruvate

- Decarboxylation step in acetyl CoA synthesis
- Pyruvate combines with ionized (carbanion) form of thiamine pyrophosphate (TPP)
- Decarboxylation yields hydroxyethyl-TPP
- Catalyzed by pyruvate dehydrogenase (E1) component of the multienzyme complex
- This is the rate-limiting step in acetyl CoA synthesis
- TPP acts as the coenzyme for E1

Pyruvate Dehydrogenase Forms Acetyl Coenzyme A from Pyruvate

- Oxidation step in acetyl CoA synthesis
- Hydroxyethyl group on TPP is oxidized to an acetyl group
- Acetyl group is transferred to lipoamide (from lipoic acid)
- Forms energy-rich thioester bond
- Lipoamide disulfide is reduced to disulfhydryl form
- Catalyzed by pyruvate dehydrogenase (E1)
- Yields acetyl–lipoamide

Pyruvate Dehydrogenase Forms Acetyl Coenzyme A from Pyruvate

- Formation of acetyl CoA
- Acetyl group transferred from acetyl–lipoamide to CoA
- Catalyzed by dihydrolipoyl transacetylase (E2)
- Thioester bond energy is preserved during transfer
- Acetyl CoA is produced—fuel for the citric acid cycle

Pyruvate Dehydrogenase Forms Acetyl Coenzyme A from Pyruvate

- Regeneration of lipoamide is required to continue the catalytic cycle
- Dihydrolipoamide must be oxidized back to lipoamide
- Catalyzed by dihydrolipoyl dehydrogenase (E3)
- Electrons transferred to FAD prosthetic group, then to NAD^+
- NADH is produced as a result

Quick Quiz 1

Acetyl CoA is:

A. the activated form of acyl groups.
B. formed by citrate synthase.
C. the fuel for the citric acid cycle.
D. A and C.
E. A, B, and C.

Answer:

How do the distinct three active site work in concert?

Flexible linkages allow lipoamide to move between different active sites.

- The three enzymes of the pyruvate dehydrogenase complex are structurally integrated, and the lipoamide arm allows rapid movement of substrates and products from one active site of the complex to another.

Analogous to biotin-binding domains.

E2 core of the Pyruvate Dehydrogenase complex. This is one subunit (out of 3) with three domains.

Reactions of the Pyruvate Dehydrogenase Complex

Figure 18.7
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Reactions of the Pyruvate Dehydrogenase Complex

1. E1 decarboxylates pyruvate, forming hydroxyethyl-TPP; CO_2 is released
2. E1 active site is deep inside the complex, connected by a 20 Å hydrophobic channel
3. E2 inserts lipoamide arm into E1's channel to access the active site
4. E1 transfers acetyl group to the lipoamide
5. Acetylated lipoamide moves to E2 active site in the E2 cube
6. Acetyl group is transferred to CoA, forming acetyl CoA (second product)
7. Reduced lipoamide arm swings to E3 active site
8. E3 oxidizes lipoamide using FAD, regenerating active lipoamide
9. NADH is produced as $FADH_2$ is reoxidized to FAD
10. Cycle is ready to begin again

The Pyruvate Dehydrogenase Complex Is Regulated by Two Mechanisms

- The formation of acetyl CoA from pyruvate is irreversible in animal cells.
- Acetyl CoA has two principle fates: metabolism by the citric acid cycle or incorporation into fatty acids. Enzyme E1 is a key site of regulation. A kinase associated with the complex phosphorylates and inactivates E1.
- A phosphatase, also associated with the complex, removes the phosphate and thereby activates the enzyme.
- The pyruvate dehydrogenase complex is also regulated by energy charge.
- ATP, acetyl CoA, and NADH inhibit the complex.
- ADP and pyruvate stimulate the complex.

Figure 18.8
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Regulation of the Pyruvate Dehydrogenase Complex

- PDH kinase phosphorylates and inactivates ***pyruvate dehydrogenase (PDH)***, and ***PDH phosphatase*** activates the dehydrogenase by removing the phosphoryl group.
- The kinase and the phosphatase are highly regulated enzymes.

Figure 18.10
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- In resting muscle, energy demand is low and there are high ratios of NADH/NAD+, acetyl CoA/CoA, and ATP/ADP
- These high ratios activate pyruvate dehydrogenase kinase
- Kinase phosphorylates and deactivates the pyruvate dehydrogenase complex
- High levels of acetyl CoA, NADH, and ATP inhibit the complex
- Pyruvate dehydrogenase is switched off when energy charge is high

Quick Quiz 2

Pyruvate dehydrogenase is _____ when ATP–ADP ratios are high.

A. activated

B. inhibited

C. phosphorylated

D. B and C

E. A and C

The Krebs Cycle/The Citric Acid Cycle (CAC)/ The Tricarboxylic Acid Cycle (TAC)

(Left) Chalermkiat Seedokmai/Getty Images.

- The Krebs cycle is considered a metabolic hub because it connects the breakdown of carbohydrates, fats, and proteins.
- It also provides key intermediates for biosynthetic pathways, allowing efficient traffic flow of energy and building blocks in the cell.

Two Stages Of the Cycle

- The citric acid cycle is the biochemical hub of the cell, oxidizing carbon fuels, usually in the form of acetyl CoA, and serving as a source of precursors for biosynthesis.
- The citric acid cycle oxidizes the acetyl fragment of acetyl CoA to CO_2.
- In the process of oxidation, high-energy electrons are captured in the form of NADH and $FADH_2$.
- The function of the citric acid cycle is to harvest high-energy electrons from carbon fuels.

Figure 19.1
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But be reminded that cellular respiration has two parts

Figure 19.2
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Stage One Oxidizes Two Carbon Atoms to Gather Energy-Rich Electrons

- The first stage generates two molecules of CO_2 by oxidative decarboxylations.
- ***Citrate synthase*** catalyzes the condensation of acetyl CoA and oxaloacetate to form citrate.

Figure 19.1
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Stage One Oxidizes Two Carbon Atoms to Gather Energy-Rich Electrons

- ***Citrate synthase*** exhibits induced fit (minimizes unwanted hydrolysis of acetyl CoA)
- Oxaloacetate binding by citrate synthase induces structural changes that lead to the formation of the acetyl CoA binding site.
- The formation of the reaction intermediate citryl CoA causes a structural change that completes active site formation.
- ***Citryl CoA*** is cleaved to form citrate and coenzyme A.

Figure 19.3
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Quick Quiz 3

Formation of citrate from acetyl CoA and oxaloacetate is a(n) _____ reaction.

A) oxidation
B) reduction
C) condensation
D) ligation
E) None of the above.

Stage One Oxidizes Two Carbon Atoms to Gather Energy-Rich Electrons

- Citrate is isomerized into isocitrate:

- Isocitrate is oxidized and decarboxylated:

Stage One Oxidizes Two Carbon Atoms to Gather Energy-Rich Electrons

***Succinyl coenzyme A* is formed by the oxidative decarboxylation of alpha-ketoglutarate.**

- Isocitrate dehydrogenase catalyzes the oxidative decarboxylation of isocitrate, forming α-ketoglutarate and capturing high-energy electrons as NADH.
- α-Ketoglutarate dehydrogenase complex catalyzes the synthesis of succinyl CoA from α-ketoglutarate, generating NADH.
- The enzyme and the reactions are structurally and mechanistically similar to the pyruvate dehydrogenase complex.

Figure 19.1
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α-Ketoglutarate **Succinyl CoA**

Stage Two Regenerates Oxaloacetate and Harvests Energy-Rich Electrons

A compound with high phosphoryl-transfer potential is generated from *succinyl coenzyme A*.

- Succinyl CoA synthetase catalyzes the cleavage of a thioester linkage and concomitantly forms ATP.

Figure 19.1
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Stage Two Regenerates Oxaloacetate and Harvests Energy-Rich Electrons

***Succinyl coenzyme A synthetase* transforms types of biochemical energy.**

- Cleavage of the thioester of succinyl CoA powers the formation of ATP.
- The formation of ATP by succinyl coenzyme A synthetase is an example of a substrate-level phosphorylation because succinyl phosphate, a high phosphoryl-transfer potential compound, donates a phosphate to ADP.

Stage Two Regenerates Oxaloacetate and Harvests Energy-Rich Electrons

Oxaloacetate is regenerated by the oxidation of succinate.

- ***Succinate dehydrogenase***, ***fumarase***, and ***malate dehydrogenase*** catalyze successive reactions to regenerate oxaloacetate.
- $FADH_2$ and NADH are generated.
- Oxaloacetate can condense with another acetyl CoA to initiate another cycle.

Figure 19.1
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Quick Quiz 4

Which of the following occurs during the citric acid cycle?

A. Generation of acetyl CoA

B. Generation of a five-carbon molecule via a condensation reaction

C. Generation of a four-carbon molecule from a five-carbon molecule via decarboxylation reaction

D. All of the above.

E. A & C

Stage Two Regenerates Oxaloacetate and Harvests Energy-Rich Electrons

$$\text{Acetyl CoA} + 3\ NAD^+ + FAD + ADP + P_i + 2\ H_2O \rightarrow$$
$$2\ CO_2 + 3\ NADH + FADH_2 + ATP + 2\ H^+ + CoA$$

The Citric Acid Cycle is Regulated

- The key control points in the citric acid cycle are the reactions catalyzed by ***isocitrate dehydragenase*** and ***α-ketoglutatrate dehydrogenase***.
- Recall that all pyruvate dehydrogenases control entry of glucose-derived acetyl CoA into cycle.

The Citric Acid Cycle is Regulated

- **The citric acid cycle must be capable of being rapidly replenished.**
- Because the citric acid cycle provides precursors for biosynthesis, reactions to replenish the cycle components are required if the energy status of the cells changes.
- These replenishing reactions are called ***anapleurotic*** reactions.
- A prominent anapleurotic reaction is catalyzed by pyruvate carboxylase. Recall that this reaction is also used in gluconeogenesis and is dependent on the presence of acetyl CoA.

$$\text{Pyruvate} + CO_2 + \text{ATP} + H_2O \longrightarrow \text{oxaloacetate} + \text{ADP} + P_i$$

The Glyoxylate Cycle Enables Plants and Bacteria to Convert Fats into Carbohydrates

- The glyoxylate cycle is similar to the citric acid cycle but bypasses the two decarboxylation steps, allowing the synthesis of carbohydrates from fats.
- Succinate can be converted into oxaloacetate and then into glucose.
- The glyoxylate cycle is prominent in oil-rich seeds such as sunflower seeds.

Quick Quiz 5

The sum reaction of the glyoxylate pathway shows that carbon enters the cycle from _____ and leaves as _____.

A. acetyl CoA; succinate

B. acetyl CoA; CO_2

C. acetyl CoA; isocitrate

D. succinate; acetyl CoA

E. oxaloacetate; citrate.

Assigned Problems

Chapter	Tymochko, Berg, Stryer, Biochemistry, 2nd Edition,	**Chapter**	Tymochko, Berg, Stryer, Biochemistry, 2nd Edition,
18	3, 6, 8, 9, 10, 12, 13, 15, 16, 18.	19	2, 4, 7, 10, 17, 19, 20, 22, 23, 24.
Chapter	Tymochko, Berg, Stryer, Biochemistry, 3nd Edition, 4th Edition (bottom line)	**Chapter**	Tymochko, Berg, Stryer, Biochemistry, 3nd Edition, 4th Edition (bottom line)
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