Chapter 16
Fatty Acid Oxidation and Ketogenesis
Fatty Acid Oxidation

• Responsible for providing energy to most cells while resting. Exceptions: brain and kidney use glucose.

• Excess acetyl-CoA is converted into fatty acids that can be stored or exported from the liver as triacylglycerols.
Why do fats have the most energy content per gram?

- Fats: 9 kcal/gram
- Sugars: 4 kcal/gram
- Amino acids: 4 kcal/gram
- Ethanol: 6 kcal/gram (for fun, calculate the calories in a rum and coke made with 30 mL of \text{rum}, 50 proof, and 210 mL of diet coke.)
Fat vs Sugar vs Amino acid

9 Cal/gram  4 Cal/gram  4 Cal/gram  6 Cal/gram

Copyright © 2017 W. W. Norton & Company
Metabolism is a combustion reaction

- Fuel is oxidized
- Oxygen is reduced
- Fuel that has more H has more e- to lose
- Which fuel has more H per C?

A. Fat
B. Sugar
C. Proteins
D. Ethanol
E. They are all the same
Key Enzymes Involved in Fatty Acid Metabolism

- Fatty acyl-CoA synthetase
  - Catalyzes priming reaction in fatty acid metabolism
  - Converts free fatty acids in the cytosol into fatty acyl-CoA
- Carnitine acyltransferase I
  - Catalyzes the rate-limiting step in fatty acid oxidation
- Acetyl-CoA carboxylase
  - Catalyzes the rate-limiting step in fatty acid synthesis
- Fatty acid synthase
  - Catalyzes a series of reactions that add C2 units to a growing fatty acid chain
Formation of Fatty Acyl-CoA

• Energetically favorable
• Fatty acyl-CoA synthetases differ in specificity by the size of the fatty acids.
• Two step reaction:
  – Fatty acid adenylation occurs (ATP coupled).
  – CoA-SH attacks adenylate intermediate and releases AMP.
Formation of Fatty Acyl-CoA

\[
\text{Adenosine} \quad \text{ATP} \quad \text{Fatty acid} \\
\text{Fatty acyl-CoA synthetase} \quad \text{Pyrophosphate} \\
\Delta G'' = -19 \text{kJ/mol} \\
\text{Pyrophosphatase} \\
\text{AMP} + \text{ATP} \rightleftharpoons \text{ADP} + \text{ADP} \\
\text{Adenylate kinase} \\
\text{ADP} + \text{ATP} \rightleftharpoons \text{ATP} + \text{ADP} \\
\text{Nucleoside diphosphate kinase} \\
\text{AMP} + 2 \text{ATP} \rightleftharpoons \text{ATP} + 2 \text{ADP} \\
\text{Net reaction} \\
\Delta G'' = -15 \text{kJ/mol} \\
\text{Fatty acyl-adenylate (enzyme-bound intermediate)} \\
\text{Fatty acyl-CoA synthetase} \\
\text{Fatty acyl-CoA}
\]
What makes the formation of fatty acyl-CoA energetically favorable?

I. Two molecule of ATP are become 2 ADP
II. The reaction of CoASH with acetyl-AMP is exergonic
III. The hydrolysis of pyrophosphate, catalyzed by pyrophosphatase is exergonic
IV. One molecule of ATP is hydrolyzed.

A. I and III  B. II and IV  C. I, II and III  D. IV only  E. none of these
What Happens to Fatty Acyl-CoA?

• If energy cell charge is low:
  – Fatty acyl-CoA is imported into the mitochondrial matrix by the carnitine transport cycle.
  – Degrades fatty acids to:
    • Acetyl-CoA
    • FADH$_2$
    • NADH

• If energy cell charge is high:
  – Fatty acid synthesis is favored.
  – Mitochondrial import of fatty acyl-CoA is inhibited by malonyl-CoA.
Carnitine Transport Cycle

• Regulates cellular metabolism by:
  – Controlling the flux of fatty acids to either degrade them (mitochondrial matrix) or synthesize them and membrane lipids (cytosol)
  – Maintains a separate pool of coenzyme A

• Controlled by malonyl-CoA
  – Inhibits carnitine acyltransferase I
  – Prevents import of fatty acyl-CoA into the mitochondria
Carnitine Transport Cycle

Figure 16.3
Biochemistry, First Edition. Copyright © 2017 W. W. Norton & Company
Fatty Acid Degradation (beta-Oxidation Reactions)

View the Fatty Acid Degradation (beta-Oxidation Reactions) animation
Fatty Acid β-oxidation Pathway

• Occurs in the mitochondria
• Degrades fatty acids C$_2$ units at a time via thiolysis
• Generates FADH$_2$, NADH, and acetyl-CoA
• Consists of four repeatable reactions
Fatty Acid β-oxidation Pathway

• Four steps:
  – Acyl-CoA dehydrogenase (oxidation)
    • Forms FADH$_2$ and a trans C─C bond
    • Isoform dependent on number of carbons in fatty acid chain
  – Enoyl-CoA hydratase (hydration)
    • Adds H$_2$O across the C═C bond
    • Stereospecific
  – 3-hydroxyacyl-CoA dehydrogenase (oxidation)
    • Forms NADH
  – B-ketoacyl-CoA thiolase (thiolysis)
    • Forms acetyl-CoA
    • Removes C$_2$ unit from the fatty acid
Fatty Acid β-oxidation Pathway

Figure 16.4

Fatty Acid β-oxidation Products and ATP Yield

Table 16.1 ATP YIELD FROM THE COMPLETE OXIDATION OF PALMITOYL-COA

<table>
<thead>
<tr>
<th>β oxidation of palmitoyl-CoA</th>
<th>Citrate cycle</th>
<th>ATP generated by oxidative phosphorylation</th>
</tr>
</thead>
<tbody>
<tr>
<td>7 NADH →</td>
<td></td>
<td>17.5 ATP</td>
</tr>
<tr>
<td>7 FADH₂ →</td>
<td></td>
<td>10.5 ATP</td>
</tr>
<tr>
<td>8 Acetyl-CoA →</td>
<td>24 NADH →</td>
<td>60 ATP</td>
</tr>
<tr>
<td></td>
<td>8 FADH₂ →</td>
<td>12 ATP</td>
</tr>
<tr>
<td></td>
<td>8 GTP →</td>
<td>8 ATP</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>108 ATP per palmitoyl-CoA</td>
</tr>
</tbody>
</table>

Note: Values for the ATP currency exchange ratio are 2.5 ATP per NADH and 1.5 ATP per FADH₂.
Electron Transfer and β-oxidation

Figure 16.6
Unsaturated fats use different enzymes

What step is skipped in B Oxidation?
What is not made?
A. NADPH
B. FADH$_2$
C. ATP
D. NADH
E. None of these

Metabolism of Polyunsaturated fats:
A. Uses ATP
B. Requires an NADPH
C. Eliminates an FADH$_2$
D. Both B and C
E. None of these
Propionoyl-CoA Forms Acetyl-CoA
Ketogenesis

• Process in which excess acetyl-CoA is converted to ketone bodies (acetoacetate and D-β-hydroxybutyrate)
• Occurs during starvation while carbohydrate sources are limited
• Ketone bodies are exported from liver into muscle tissues.
Ketogenesis

Figure 16.14

Copyright © 2017 W. W. Norton & Company
Formation of Ketone Bodies

\[
\text{Acetyl-CoA} + \text{Acetyl-CoA} \xrightarrow{\beta\text{-Ketoacyl-CoA thiolase}} \text{Acetoacetyl-CoA} \xrightarrow{\text{HMG-CoA synthase}} \text{3-Hydroxy-3-methylglutaryl-CoA (HMG-CoA)} \xrightarrow{\text{HMG-CoA lyase}} 2 \text{Acetyl-CoA}
\]
Figure 16.15

3-Hydroxy-3-methylglutaryl-CoA (HMG-CoA) is converted to Acetyl-CoA by HMG-CoA lyase. Acetyl-CoA is then converted to Acetoacetate by Acetoacetate decarboxylase, which releases CO₂. Acetoacetate is then converted to Acetone by the action of d-β-Hydroxybutyrate dehydrogenase. The d-β-Hydroxybutyrate is exported to the blood.

In the tissues (but not the liver), ketone bodies are converted to acetyl CoA.
16.2 Synthesis of Fatty Acids and Triacylglycerols

• Carbon substrates are primarily derived from dietary carbohydrates (i.e., glucose).
Carbohydrate and Fatty Acid Metabolism

View the Carbohydrate and Fatty Acid Metabolism animation
Differences Between Fatty Acid Degradation and Synthesis

**Mitochondrial matrix**

Fatty acid degradation

1. Oxidation
2. Hydration
3. Oxidation
4. Cleavage

Acetyl-CoA

- CoA dependent
- FAD/NAD⁺ dependent
- Multiple enzymes
- Carnitine transport is rate limiting

**Cytosol**

Fatty acid synthesis

4. Reduction
3. Dehydration
2. Reduction
1. Condensation

Malonyl-CoA

Acetyl-CoA + CO₂

- ACP dependent
- NADPH dependent
- Two enzymes
- Malonyl-CoA synthesis is rate limiting

**Figure 16.18**
Fatty Acid Synthesis – Part 1

• Multifunctional enzyme
  – Uses acyl carrier protein as a hydrocarbon anchor
  – Rate-limiting step is the generation of malonyl CoA by acetyl-CoA carboxylase.

• Four step reaction
  – $\beta$-ketoacyl-ACP synthase (condensation)
  – $\beta$-ketoacyl-ACP reductase (reduction)
  – $\beta$-ketoacyl-ACP dehydratase (dehydration)
  – Enoyl-ACP reductase (reduction)
Fatty Acid Synthesis – Part 2


2. Reduction of \(\beta\)-Ketoacyl-ACP to \(\beta\)-Hydroxybutyryl-ACP by \(\beta\)-Ketoacyl-ACP reductase (KR) using NADPH.

3. Dehydration of \(\beta\)-Hydroxybutyryl-ACP to Butenoyl-ACP by \(\beta\)-Hydroxyacyl-ACP dehydratase (DH).

4. Reduction of Butenoyl-ACP to Butyryl-ACP by Enoyl-ACP reductase (ER) using NADPH.

Figure 16.21
Acetyl-CoA Carboxylase

Figure 16.19
Acyl Carrier Protein Structure

Phosphopantetheine group

Hexanoyl-ACP

Acetyl-CoA

Figure 16.22a
Fatty Acid Synthesis (Fatty Acid Synthase Reactions)

View the Fatty Acid Synthesis (Fatty Acid Synthase Reactions) animation
Fatty Acid Synthesis of Palmitate

- $8 \text{ Acetyl-CoA} + 7 \text{ ATP} + 14 \text{ NADPH} + 14 \text{ H}^+ \rightarrow$
- Palmitate + $8 \text{ CoA} + 7 \text{ ADP} + 7 \text{ Pi} + 14 \text{ NADP}^+ + 6 \text{ H}_2\text{O}$
Elongation Enzymes

- Used to increase carbon chain in palmitate to make longer fatty acids
Desaturating Enzymes

- Membrane-bound ER proteins that use $\text{O}_2$ as an oxidant
- Produces unsaturated fatty acids
Triacylglycerol Synthesis

- Formed from phosphatidic acid
- Dephosphorylation occurs
- Addition of fatty acids through esterification
Formation of Phospholipids

Figure 16.29
Formation of Sphingolipids – Part 1

Figure 16.30


Copyright © 2017 W. W. Norton & Company
Formation of Sphingolipids – Part 2

Figure 16.31


Copyright © 2017 W. W. Norton & Company
Regulation of Fatty Acid Synthesis – Part 1

• Modulation of acetyl-CoA carboxylase activity is controlled by:
  – Allosteric mechanisms
    – Citrate and palmitoyl-CoA
  – Covalent modification
    • AMP-activated protein kinase (AMPK)
Regulation of Fatty Acid Synthesis – Part 2

Figure 16.33

AMPK Mechanism

AMP-activated protein kinase

Inactive AMPK

Partialy active AMPK

AMP kinase (LKB1)

Active acetyl-CoA carboxylase polymer

Inactive acetyl-CoA carboxylase monomer

Fully active AMPK

High levels of malonyl-CoA and increased flux through the fatty acid synthesis pathway

Low levels of malonyl-CoA and decreased flux through the fatty acid synthesis pathway

AMP

ATP

ADP

PPi

H2O

Insulin

Protein phosphatase 2C

Copyright © 2017 W. W. Norton & Company
Flux Through Fatty Synthesis and Degradation

Figure 16.35
Biochemistry, First Edition, Copyright © 2017 W. W. Norton & Company
16.3 Cholesterol Synthesis and Metabolism

• Cholesterol synthesis occurs in all cells but primarily occurs in the liver.

• Consists of 4 distinct stages:
  – Formation of mevalonate from acetyl-CoA
  – Formation of isopentenyl diphosphate
  – Formation of squalene
  – Cyclization of squalene
Cholesterol

- Plays a critical role in cell membrane function and production of cell signaling molecules
Cholesterol Synthesis Overview

Figure 16.36
Cholesterol Synthesis – Stage 1

- 2 molecules of acetyl-CoA are condensed.
- HMG-CoA reductase is the rate limiting step of the entire pathway.
- Mevalonate is produced.
Cholesterol Synthesis – Stage 2

- ATP donates 2 phosphoryl groups.
- Isoprene units are involved.
- Dimethylallyl diphosphate is produced.

![Diagram of cholesterol biosynthesis stage 2]
Cholesterol Synthesis – Stage 3 – Part 1

- Isoprene (C$_5$) units are attached to form a geranyl diphosphate (C$_{10}$) compound.
- Farnesyl diphosphate (C$_{15}$) is made.
- Formation of squalene
Cholesterol Synthesis – Stage 3 – Part 2

Figure 16.39
Cholesterol Synthesis – Stage 4

- Squalene (C_{30}) is cyclized.
- Lanosterol is converted into cholesterol in 19 steps.
Fates of Cholesterol from the Liver – Part 1

• Three major functions:
  1. Stored in intracellular lipid droplets
  2. Packaged into lipoproteins and exported into circulatory system
  3. Secreted into small intestines through the bile duct
Fates of Cholesterol from the Liver – Part 2

Figure 16.41
Cholesterol Metabolism and Cardiovascular Disease

• Statin drugs can be used to decrease the risk of cardiovascular disease.
Lipoproteins

• Apolipoproteins – membrane-bound vesicles contain a hydrophobic core and one or more proteins on the surface
• Consist of a phospholipid monolayer containing cholesterol and one or more apolipoproteins
• Serve as signaling molecules
• Differ depending on protein:triglyceride ratio and densities
# Lipoprotein Classes

## Table 16.2 MAJOR CLASSES OF LIPOPROTEINS IN HUMAN SERUM

<table>
<thead>
<tr>
<th>Feature</th>
<th>Chylomicron</th>
<th>Very-low-density lipoprotein</th>
<th>Intermediate-density lipoprotein</th>
<th>Low-density lipoprotein</th>
<th>High-density lipoprotein</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diameter (nm)</td>
<td>100–1,000</td>
<td>30–80</td>
<td>25–35</td>
<td>18–25</td>
<td>5–12</td>
</tr>
<tr>
<td>Density (g/cm$^3$)</td>
<td>&lt;0.95</td>
<td>&lt;1.006</td>
<td>&lt;1.006–1.019</td>
<td>&lt;1.019–1.063</td>
<td>&lt;1.063–1.210</td>
</tr>
<tr>
<td>Percent protein (%)</td>
<td>2</td>
<td>10</td>
<td>20</td>
<td>25</td>
<td>55</td>
</tr>
<tr>
<td>Percent triacylglycerol (%)</td>
<td>85</td>
<td>60</td>
<td>22</td>
<td>10</td>
<td>5</td>
</tr>
<tr>
<td>Percent cholesterol ester (%)</td>
<td>5</td>
<td>15</td>
<td>30</td>
<td>40</td>
<td>12</td>
</tr>
<tr>
<td>Major apolipoproteins</td>
<td>apoC-II, apoC-III, apoB-48, apoE</td>
<td>apoC-II, apoC-III, apoB-100, apoE</td>
<td>apoC-II, apoC-III, apoB-100, apoE</td>
<td>apoB-100</td>
<td>apoC-II, apoC-III, apoA-I, apoA-II, apoD</td>
</tr>
</tbody>
</table>

*Note: apo = apolipoprotein.*

Lipoprotein Structures

- Multiple classes including:
  - Chylomicrons
  - VLDL
  - IDL
  - Chylomicron remnants
  - LDLs
HDL Particles Reverse Cholesterol Transport

- HDL particles remove cholesterol from peripheral tissues through apoA-I.
- Cholesterol from peripheral tissue is taken back into the liver.
Cholesterol Homeostasis

Figure 16.50

Cholesterol-Lowering Drugs

Simvastatin (Zocor)

Lovastatin (Mevacor)

HMG-CoA

Rosuvastatin (Crestor)

Atorvastatin (Lipitor)

Figure 16.51

SREBP

• Large proteins embedded in the ER membrane
• Low levels of intracellular cholesterol stimulate binding of SREBPs to SRE sequences located in the transcriptional control region of specific genes
• Control mechanism for cholesterol biosynthesis
• SREs have a high affinity binding site for SREBP.
SREBP Binding

Figure 16.57

Copyright © 2017 W. W. Norton & Company
Clicker Question 1

• Myristic acid (C\textsubscript{14}) is completely metabolized via β-oxidation. Which products are formed?

  – 6 NADH, 6 FADH\textsubscript{2}, 6 acetyl-CoA
  – 6 NADH, 6 FADH\textsubscript{2}, 7 acetyl-CoA
  – 7 NADH, 7 FADH\textsubscript{2}, 6 acetyl-CoA
  – 7 NADH, 7 FADH\textsubscript{2}, 7 acetyl-CoA
Clicker Question 1 – Answer

• Myristic acid \((C_{14})\) is completely metabolized via \(\beta\)-oxidation. Which products are formed?

b. 6 NADH, 6 FADH\(_2\), 7 acetyl-CoA
Clicker Question 2

• Arachidonic acid (C$_{20}$) is fully metabolized by fatty acid degradation. How many rounds does it take for this to happen?

- 7
- 8
- 9
- 10
Clicker Question 2 – Answer

• Arachidonic acid ($C_{20}$) is fully metabolized by fatty acid degradation. How many rounds does it take for this to happen?

c.9
Clicker Question 3

• Ketogenesis produces which products?

a. Acetyl-CoA
b. Acetoacetate
c. D-β-hydroxybutyrate
d. A and B
e. B and C
Clicker Question 3 – Answer

• Ketogenesis produces which products?

e. B and C
Clicker Question 4

• When acetyl CoA levels are too high, they are exported through the __________ in the mitochondria.

a. acetyl-CoA transporter
b. β-oxidation pathway
c. citrate shuttle
d. glycolytic pathway
e. malate shuttle
Clicker Question 4 – Answer

• When acetyl CoA levels are too high, they are exported through the _________ in the mitochondria.

c. citrate shuttle
Clicker Question 5

• Statin drugs inhibit which stage of cholesterol biosynthesis?

  a. Stage 1
  b. Stage 2
  c. Stage 3
  d. Stage 4
  e. Stage 5
Clicker Question 5 – Answer

• Statin drugs inhibit which stage of cholesterol biosynthesis?

a. Stage 1
This concludes the Lecture PowerPoint presentation for Chapter 16