Lipid metabolism

- Degradation and biosynthesis of fatty acids
- Ketone bodies
Fatty acids (FA)

- primary fuel molecules in the fat category
- main use is for long-term energy storage
- high level of energy storage: fats - 38 kJ/g, carbohydrates 16 kJ/g

Primary sources of FA:
- dietary triacylglycerols
- triacylglycerols synthesized in the liver
- triacylglycerols stored in adipocytes

FA must be delivered to cells where β-oxidation occurs:

FA → cell → β-oxidation → ATP
  • liver
  • heart
  • skeletal muscles
Fatty acid degradation - β-oxidation

- FA are delivered to cells by diffusion from blood capillaries
- upon entry into the cell FA are immediately activated by linking them as thioesters to CoASH
- activation is coupled with FA transport through the mitochondrial membrane

**β-oxidation**: two-carbon fragments successively removed from the carboxyl end of activated FA producing acetyl-CoA → entry into TCA cycle → ATP

*lokализованный в митохондриальной матрице* (>18 C peroxisomes)
Fatty acid degradation

FA $\rightarrow$ entry into the cell (cytosol) $\rightarrow$ acyl-CoA $\rightarrow$ transport across the inner mitochondrial membrane (carnitine shuttle)

$\rightarrow$ acyl-CoA $\rightarrow$ $\beta$-oxidase $\rightarrow$ acetyl-CoA $\rightarrow$ TCA cycle

Fatty acid activation – formation of acyl–CoA:

Fatty acyl coenzyme A-synthetase
**Carnitine shuttle** – transport of long-chain FA (>12C) into the mitochondrial matrix

- **CAT** – carnitine acyltransferase (carnitine palmitoyltransferase)
- **Carnitine-acylcarnitine translocase**
FA activation and transport across inner mitochondrial membrane
β-Oxidation of fatty acids – spiral pathway

Thiolytic cleavage

Dehydrogenation/oxidation

Hydration

Dehydrogenation/oxidation

ET chain 2 ATP

ET chain 3 ATP
Combination of FA activation, transport into the mitochondrial matrix, β-oxidation and TCA cycle
Energy yield from β-oxidation of saturated FA

example: stearic acid, 18 C → 9 acetyl-Coa, 8 turns of β-oxidation

<table>
<thead>
<tr>
<th>ATP/unit</th>
<th>ATP produced</th>
</tr>
</thead>
<tbody>
<tr>
<td>activation →</td>
<td>-1(-2)</td>
</tr>
<tr>
<td>9 acetyl CoA</td>
<td>TCA</td>
</tr>
<tr>
<td>8 FADH$_2$</td>
<td></td>
</tr>
<tr>
<td>8 (NADH + H$^+$)</td>
<td></td>
</tr>
<tr>
<td>total ATP</td>
<td></td>
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</tbody>
</table>
β-oxidation of unsaturated fatty acids

requires additional enzymes – isomerase, NADPH-dependent reductase (2,4-dienoyl-CoA reductase), epimerase
Example: **Oleic acid**

- cis-double bond
- Δ³ cis → Δ² trans
- 3 turns of β-oxidation
- hydration → dehydrogenation → ....

Unsaturated FA provide less energy than saturated FA – less reducing equivalents (FADH₂) are produced.
**β-oxidation of FA with an odd number of carbon**

They can be handled normally until the last step where propionyl CoA is produced.

Three reactions are required to convert propionyl CoA to succinyl CoA which can enter to the TCA cycle.
Ketone bodies

- After degradation of a fatty acid, acetyl CoA is further oxidized in the citric acid cycle.

- **First step in the citric acid cycle**

  
  - acetyl CoA + oxaloacetate → citrate

- If too much acetyl CoA is produced from β-oxidation, some is converted to ketone bodies.
**Ketone bodies**

- **synthesis**: ketogenesis
- **localization**: liver, mitochondrial matrix
- **initial substrate**: acetyl-CoA
- **function**: energy source
  - physiological conditions - myocard
  - starvation - skeletal muscles
  - brain

\[
\begin{align*}
\text{acacetate} & : \text{CH}_3-\text{C}-\text{CH}_2-\text{COO}^- \\
\text{β-hydroxybutyrate} & : \text{CH}_3-\text{CH}-\text{CH}_2-\text{COO}^- \\
\text{acetone} & : \text{O} \quad \text{CH}_3-\text{C}-\text{CH}_3
\end{align*}
\]
Formation and utilization of ketone bodies

enters into TCA cycle $\rightarrow$ ATP
Ketogenesis

Liver (mitochondrial matrix)

\[
\text{CH}_3\text{-C- S-CoA} \rightarrow \text{acetoacetyl-CoA thiolase}
\]

\[
\text{O} \quad \text{CH}_3\text{-C- S-CoA} \rightarrow \text{acetoacetyl-CoA}
\]

\[
\text{O} \quad \beta\text{-hydroxy-}\beta\text{-methylglutaryl-CoA (HMG-CoA)} \rightarrow \text{HMG-CoA synthase}
\]

\[
\text{CH}_3\text{-C-CH}_2\text{-C-S-CoA} \rightarrow \text{HMG-CoA lyase}
\]

\[
\text{CH}_3\text{-C-CH}_2\text{-C-S-CoA} \rightarrow \text{acetoacetate}
\]

\[
\text{O} \quad \text{O} \quad \text{CH}_3\text{-C-CH}_2\text{-C-S-CoA} \rightarrow \text{NADH}+\text{H}^+ \rightarrow \text{NAD}^+
\]

\[
\text{CO}_2 \quad \text{acetone}
\]

\[
\text{O} \quad \text{CH}_3\text{-C-CH}_3 \rightarrow \beta\text{-hydroxybutyrate}
\]
Starvation and ketone bodies

**Blood Concentration (millimolar)**

- **hydroxybutyrate**
- **acetoacetate**

**Weeks of Starvation**

The graph shows the increase in blood concentrations of hydroxybutyrate and acetoacetate over weeks of starvation.
Fate of ketone bodies

- **Acetoacetate and β-hydroxybutyrate**
  
  Produced in the liver, diffuse in blood to other tissues.
  
  Primarily transported to muscles and brain for use as energy sources after reconversion to acetyl CoA

- **Acetone**
  
  Only produced in small amounts, eliminated in urine or breath.

\[
\begin{align*}
\text{Acetoacetate} & \quad \xrightarrow{H^+} \quad \text{Acetone}\\
\text{CO}_2 &
\end{align*}
\]

Spontaneous loss of CO\(_2\) from acetoacetate. It can be detected in the breath - *acetone breath*

Symptom of untreated diabetes mellitus or starvation conditions.
Utilization of ketone bodies in extrahepatic tissues

\[
\text{CH}_3\text{-CH-CH}_2\text{-COO}^- + \text{NAD}^+ \rightarrow \text{CH}_3\text{-CH-CH}_2\text{-COO}^- + \text{NADH} + \text{H}^+
\]

\[\beta\text{-hydroxybutyrate dehydrogenase}\]

\[
\text{CH}_3\text{-CH-CH}_2\text{-COO}^- \rightarrow \text{CH}_3\text{-CH-CH}_2\text{-C-\text{O}^-}
\]

\[\beta\text{-ketoacyl-CoA transferase}\]

\[
\text{CH}_3\text{-CH-CH}_2\text{-C-\text{O}^-} \rightarrow \text{CH}_3\text{-C-CH}_2\text{-C-\text{S-CoA}} + \text{CoASH}
\]

\[\text{acetoacetyl-CoA thiolase}\]

\[
\text{CH}_3\text{-C-CH}_2\text{-C-\text{S-CoA}} \rightarrow 2 \text{CH}_3\text{-C-\text{S-CoA}} + \text{CoASH}
\]

\[\text{acetoacetyl-CoA}\]

\[\text{acetyl-CoA}\]

\[\text{myokard at physiological conditions}\]

\[\text{skeletal muscles, brain in starvation}\]
Utilization of ketone bodies produced in the liver

\[ \text{Beta oxidation} \rightarrow 2 \text{ Acetyl CoA} \rightarrow \text{Acetoacetate} \rightarrow 3-\text{Hydroxy-3-methylglutaryl CoA (HMG CoA)} \]

\[ \text{Acetone (smell in breath)} \]

\[ \text{NADH} \rightarrow \text{Acetyl CoA} \rightarrow \text{3-hydroxybutyrate} \rightarrow \text{Acetone} \]

\[ \text{NAD}^{+} \rightarrow \text{LIVER} \]

\[ \text{MUSCLE, HEART, (BRAIN)} \]

\[ \text{Succinyl CoA} \rightarrow \text{Succinate} \rightarrow \text{Fumarate} \rightarrow \text{Malate} \rightarrow \text{Oxaloacetate} \rightarrow \text{Citrate} \rightarrow \text{Isocitrate} \rightarrow \text{Citric acid cycle} \]

\[ \text{Alpha ketoglutarate} \]
Abnormal production of ketone bodies: starvation, I. type diabetes mellitus, alcoholism

secretion of glucagon → activation of hormone sensitive lipase in adipocyte → increased lipolysis in adipose tissue → increased entry of FA into the liver → β-oxidation

high production of acetyl-CoA

+ lack of oxaloacetate

ketogenesis → ketosis → ketoacidosis

ketonemia, ketonuria
Ketone bodies – excretion from the organism

- Acetacetate
- Acetone
- β-Hydroxybutyrate

**Excretion in:**
- Breath
- Plasma
- Urine

**Percentage:**
- Acetacetate: 20%
- Acetone: 2%
- β-Hydroxybutyrate: 78%

**Ketoacidosis:**
- $\text{CH}_3\text{-C-CH}_2\text{-COO}^- + \text{H}^+ \rightarrow \text{CH}_3\text{-CH-CH}_2\text{-COO}^- + \text{H}^+ + \text{H}_2\text{O}$

**Increased loss:**
- Increased water loss
- Loss of Na$^+$
Biosynthesis of fatty acids

initial substrate: acetyl-CoA

tissues:
   liver
   adipose tissue
   lactating mammary gland

intracellular localization:
   cytosol - palmitic acid (C16)
   ER, mitochondria - elongation (C18 - C24)
   ER - desaturation – palmitooleic acid (C16)
   - oleic acid (C18)
   - arachidonic acid (C20)

Reactions of FA biosynthesis - reversal course of FA β-oxidation
β-Oxidation

\[
\begin{align*}
R- & \text{CH}_2 - \text{CH}_2 - \text{CO} - \text{S} - \\
\text{dehydrogenation} & \\
R- & \text{CH} = \text{CH}_2 - \text{CO} - \text{S} - \\
\text{hydration} & + \text{H}_2\text{O} \quad - \text{H}_2\text{O} \\
R- & \text{CH(OH)} - \text{CH}_2 - \text{CO} - \text{S} - \\
\text{dehydrogenation} & \\
R - & \text{CO} - \text{CH}_2 - \text{CO} - \text{S} - \\
\text{thiolytic cleavage} & \\
R - & \text{CO} - \text{S} - \\
\text{condensation} & + \text{CH}_3\text{- CO} - \text{SCoA}
\end{align*}
\]

Biosynthesis
Differences between FA synthesis and degradation

- **Intracellular localization**
  - degradation (β-oxidation) - mitochondrial matrix
  - biosynthesis - cytosol

- **Activation of an acyl**
  - degradation (β-oxidation) - CoA-SH
  - biosynthesis - ACP-SH (acyl carrier protein, prosthetic group phosphopantetheine)

- **Oxidoreduction cofactors**
  - degradation (β-oxidation) - NAD⁺, FAD
  - biosynthesis - NADPH + H⁺

- **Biosynthesis**
  - growth of an FA chain catalyzed by 1 multifunctional enzyme (contains aktive sites for all reactions of the synthesis) – fatty acid synthase

1. **reaction** of synthesis **distinct** from a reversal reaction of degradation

\[\text{malonyl CoA + acetyl CoA} \times \text{acetyl CoA + acetyl CoA}\]

 donor of two-carbon unit in each turn
Production of cytosolic acetyl-CoA
Transport of acetyl-CoA from mitochondrial matrix to cytosol in the form of citrate

Sources of cytosolic NADPH
- pentose phosphate pathway – oxidative phase
- oxidation of malate – malate dehydrogenase dekarboxylating (malic enzyme):

NADP⁺ → NADPH⁺ H⁺

oxalacetate → malate → pyruvate + CO₂

at high concentration of mitochondrial citrate, isocitrate dehydrogenase inhibited by ATP
Tvorba malonyl-CoA
Two-carbon units are incorporated into a growing chain in the form of malonyl-CoA

\[ \text{acetyl-CoA} \xrightarrow{\text{acetyl-CoA carboxylase}} \text{malonyl-CoA} \]

Regulation:
- Allosteric:
  - + citrate
  - - palmitoyl-CoA (product)

- Hormonal:
  - + insulin
  - - glucagon
FA biosynthesis - synthesis of palmitate by the fatty acid synthase complex

1. two-carbon unit

source of other two-carbon units for chain growth
Regulation of fatty acid metabolism

• Supply of fatty acids
  - liver
  - FA
  - β-oxidation
  at oxalacetate
  - citrate
  - ketogenesis
  adipocyte
  - FA
  - TAG synthesis

• ATP → citrate
  (glucose supply – after meal, inhibited isocitrate dehydrogenase in TCA cycle) – after translocation to cytosol
  citrate activates acetyl CoA carboxylase
  - FA synthesis

• Malonyl-CoA
  - inhibits carnitine acyltransferase I
  - long-chain FA cannot enter into mitochondria
  - FA accumulate in cytosol
  - β-oxidation
  - TAG synthesis

• Palmitoyl-CoA
  - inhibits mitochondrial translocase for citrate
  do not enter into cytosol
  - FA synthesis
Regulation of fatty acid metabolism

Hormonal regulation:

- **Insulin** - dephosphorylation of acetyl CoA carboxylase (= activation) → FA synthesis

- **Glukagon** - phosphorylation of acetyl CoA carboxylase (= inaktivace) → FA synthesis
  - increased lipolysis in adipose tissue → entry of FA into liver → β-oxidation

- **Adaptive control** - changes in enzyme expression
  - food supply after fasting - expression of acetyl CoA carboxylase, fatty acid synthase → FA synthesis
Overview of fatty acid metabolism

**TAG** - adipose tissue

Liver → **TAG** → VLDL → **LPL** → chylomicrons ← **TAG** ← GIT

**TAG** stored in tissues

complex lipids

cellular membranes

catabolism of carbohydrates and amino acids

**FA** → acetyl-CoA → HMG CoA → NADH+H⁺, FADH₂

TCA cycle

β-oxidace → biosyntéza

eicosanoids

ketone bodies

steroids

respiratory chain

ATP