Lipid metabolism - 1. Catabolism of triacylglycerols, oxidation of fatty acids and glycerol. Ketogenesis.

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Lecture plan

Catabolism of triacylglycerols in adipocytes of adipose tissue.

Biosynthesis of triacylglycerols.

Oxidation of glycerol: enzyme reactions, bioenergetics.

Lipoproteins.

Hyperlipoproteinemias.

Oxidation of glycerol: enzyme reactions, bioenergetics.

Oxidation of fatty acids (β-oxidation).

Ketone bodies.

Biosynthesis of fatty acids.

Lipids are organic compounds, are nonpolar molecules, which are soluble only in nonpolar solvents and insoluble in water.

Types of Lipids:

Simple Lipids

Esters of fatty acids with various alcohols. **Fats:** Esters of fatty acids with glycerol. Oils are fats in the liquid state

Steroids (cholesterol)

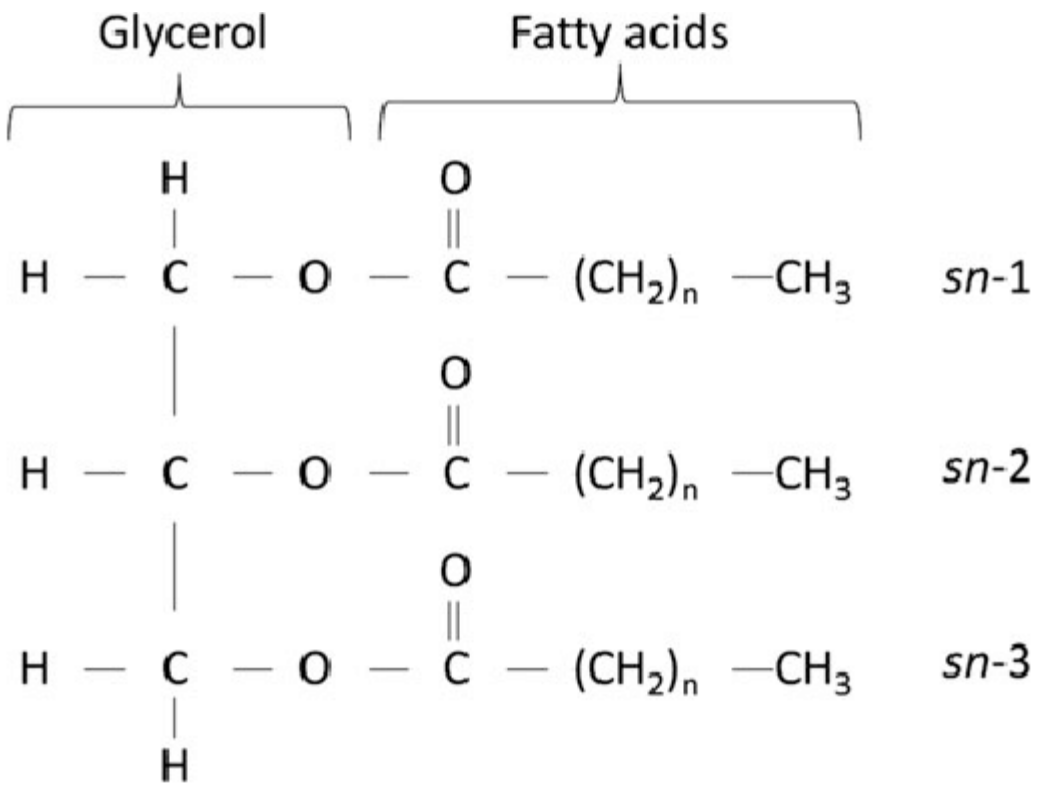
Complex Lipids

Esters of fatty acids containing groups in addition to alcohol and a fatty acid.

Phospholipids: These are lipids containing, in addition to fatty acids and alcohol, a phosphoric acid residue. They frequently have nitrogen-containing bases and other substituents, eg, in glycerophospholipids the alcohol is glycerol and in sphingophospholipids the alcohol is sphingosine.

Glycolipids (glycosphingolipids): Lipids containing a fatty acid, sphingosine, and carbohydrate.

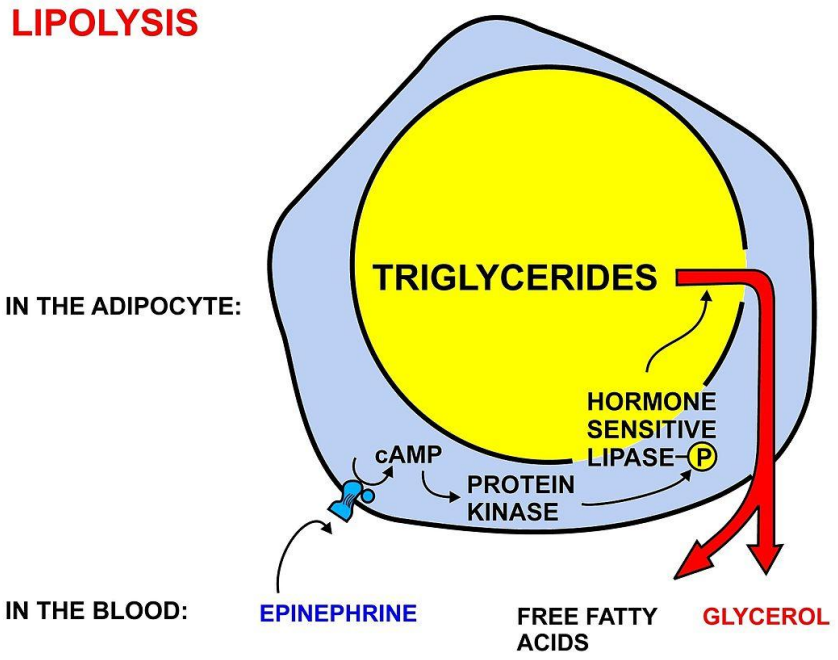
Structure of triacylglycerol



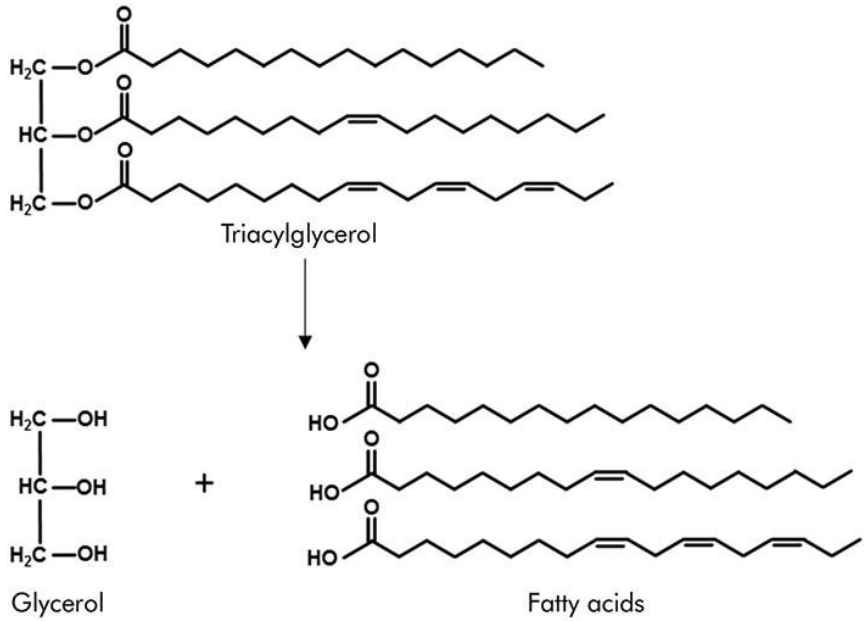
https://www.researchgate.net/figure/Triacylglycerol-TAG-structure-showing-glycerol-with-three-fatty-acids_fig1_316787887

Lipolysis is the metabolic pathway through which lipid triacylglycerols are hydrolyzed into a glycerol and three fatty acids.

Occurs in adipocytes. Lipolysis is induced by glucagon, epinephrine, norepinephrine, growth hormone, cortisol. Lipolysis is activated when: under normal physiological stressful situations - emotional stress, muscle work, fasting,
in pathological conditions - type I diabetes mellitus, other hormonal diseases (hypercortisolism, hyperthyroidism).

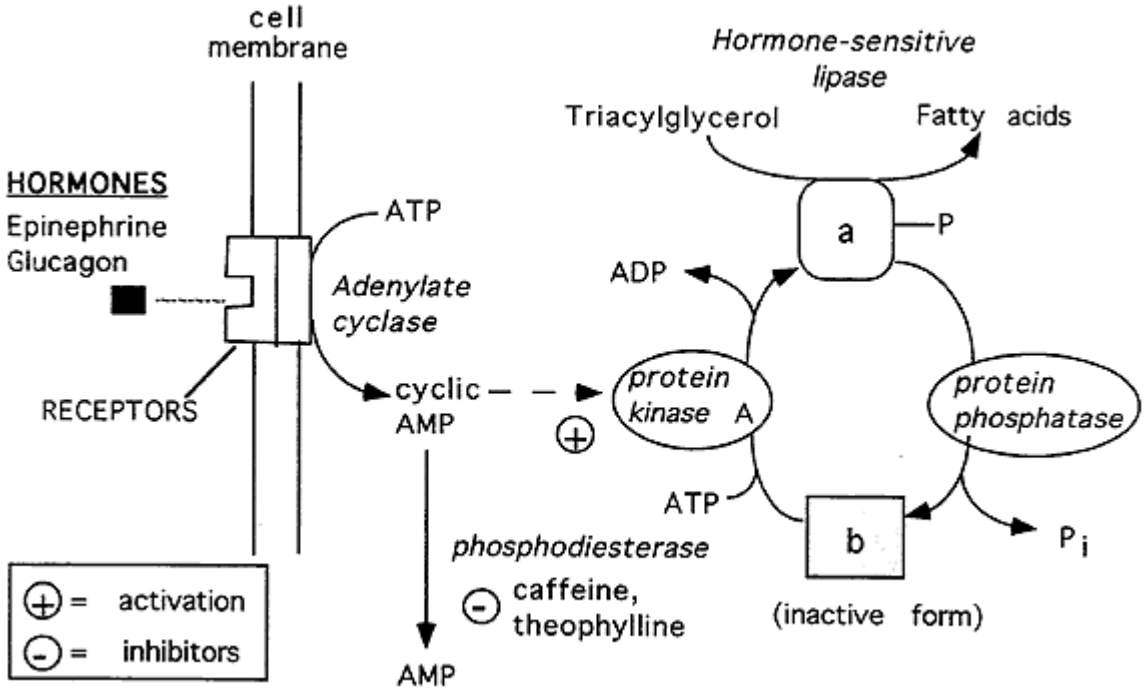


https://en.wikipedia.org/wiki/Lipolysis



https://www.researchgate.net/figure/Triacylglycerol-TAG-structure-showing-glycerol-with-three-fatty-acids_fig1_316787887

Lipolysis regulation



http://www2.csudh.edu/nsturm/CHE452/09_Fat%20Met-RegIntegr.htm

Lipogenesis

Fats not only obtained from the diet but also obtained from lipogenesis in the body. **Lipogenesis** means synthesis of neutral fats (TAG) from CHO and proteins present in excess of body need.

Lipogenesis requires: 1- Synthesis of fatty acids (FA) and glycerol
2- Activation of fatty acids by CoA and glycerol by glycerokinase,
3- the combination of activated fatty acids and activated glycerol

De *no vo* synthesis of fatty acid (cytoplasmic synthesis):

Occur mainly for the synthesis of palmitic acid

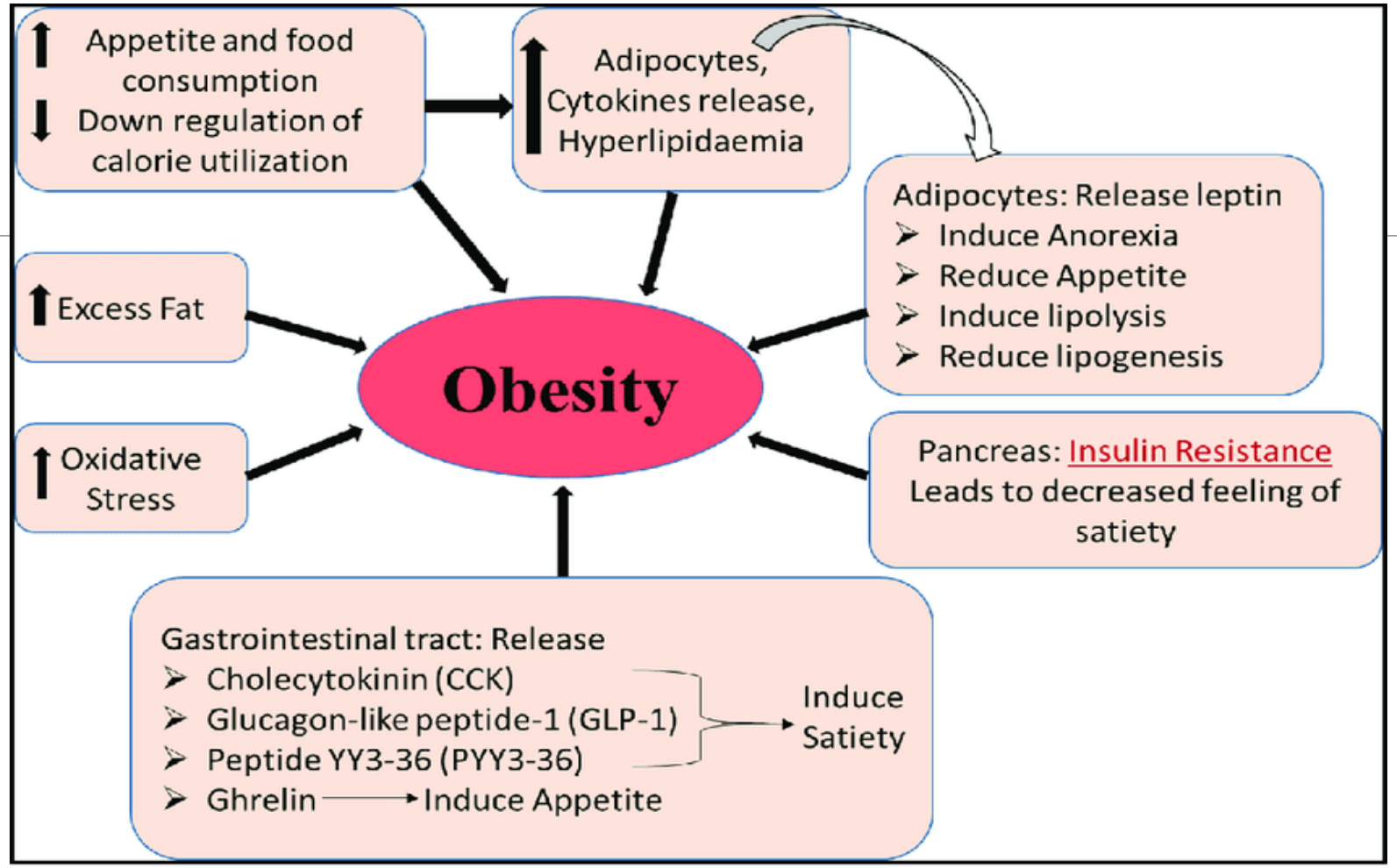
Site: ***Cytoplasm*** of liver, mammary glands and adipose tissues.

https://www.slideserve.com/castor-mills/lipogenesis

Obesity

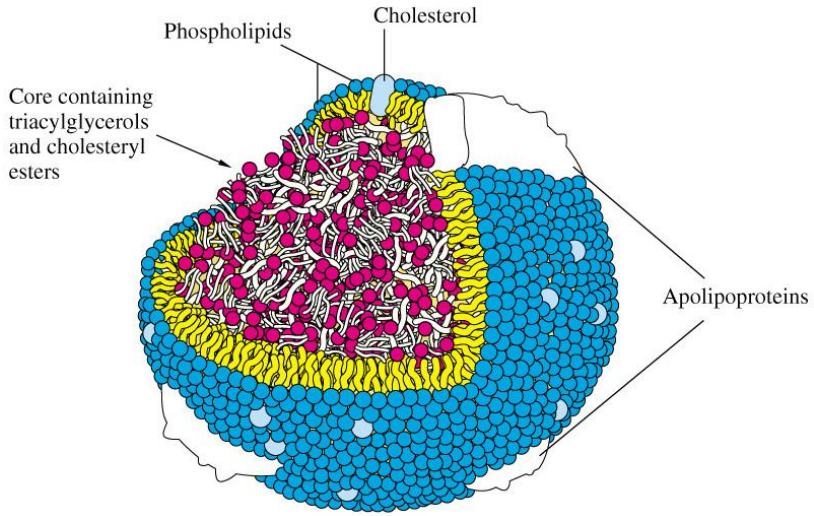
Obesity is essentially an excessive accumulation of triacylglycerols in fatty tissue that is the net result of excessive energy intake compared to energy usage.
Obesity is diagnosed when your body mass index (BMI) is 30 or higher.
To determine your body mass index, divide your weight in pounds by your height in inches squared and multiply by 703.
Or divide your weight in kilograms by your height in meters squared.

BMI	Weight status
Below 18.5	Underweight
18.5-24.9	Normal
25.0-29.9	Overweight
30.0 and higher	Obesity

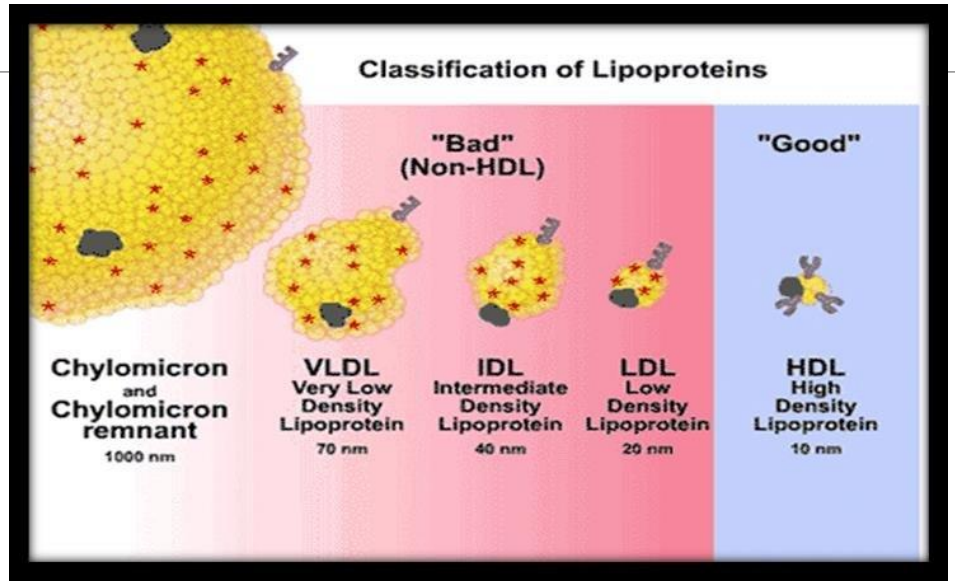


https://www.researchgate.net/figure/The-diagrammatic-representation-of-basic-pathogenesis-of-obesity_fig2_329208017

Lipoproteins



https://www.apsubiology.org/anatomy/2020/2020_Exam_Reviews/Exam_1/CH18_Lipoproteins.htm



https://www.researchgate.net/figure/4-The-major-types-of-lipoproteins-are-chylomicrons-very-low-density-lipoprotein-VLDL_fig4_304525065

Apolipoproteins

Apolipoproteins, also referred to as **apoproteins**, form the protein component of the lipoproteins described above. Apolipoproteins are receptor molecules and are involved in signaling. While it is highly unlikely that specific functions of each apolipoprotein will be tested on the MCAT, they are briefly summarized below to illustrate their diverse purposes:

- **apoA-I**: activates LCAT, an enzyme that catalyzes cholesterol esterification
- **apoB-48**: mediates chylomicron secretion
- **apoB-100**: permits uptake of LDL by the liver
- **apoC-II**: activates lipoprotein lipase
- **apoE**: permits uptake of chylomicron remnants and VLDL by the liver

Classes of lipoproteins

oprotein	Functions
Chylomicrons	Transport dietary triacylglycerols and cholesterol from intestine to tissues
VLDL	Transports triacylglycerols from liver to tissues
IDL (VLDL remnants)	Picks up cholesterol from HDL to become LDL Picked up by the liver
LDL	Delivers cholesterol into cells
HDL	Picks up cholesterol accumulating in blood vessels Delivers cholesterol to liver and steroidogenic tissues Transfers apolipoproteins to other lipoproteins

https://schoolbag.info/chemistry/mcat_biochemistry/70.html#:~:text=Chylomicrons%20are%20the%20least%20dense,HDL%20(high-density).

Chylomicrons are highly soluble in both lymphatic fluid and blood and function in the transport of dietary triacylglycerol, cholesterol, and cholesteryl esters to other tissues. Assembly of chylomicrons occurs in the intestinal lining and results in a nascent chylomicron that contains lipids and apolipoproteins.

VLDL (VERY-LOW-DENSITY LIPOPROTEIN)

VLDL metabolism is similar to that of chylomicrons; however, VLDL is produced and assembled in liver cells. Like chylomicrons, the main function of VLDL is the transport of triacylglycerol to other tissues. VLDLs also contain fatty acids that are synthesized from excess glucose or retrieved from chylomicron remnants.

IDL (INTERMEDIATE-DENSITY LIPOPROTEIN)

Once triacylglycerol is removed from VLDL, the resulting particle is referred to as either a **VLDL remnant** or **IDL**. Some IDL is reabsorbed by the liver by apolipoproteins on its exterior, and some is further processed in the bloodstream. For example, some IDL picks up cholesteryl esters from HDL to become LDL. IDL thus exists as a transition particle between triacylglycerol transport (associated with chylomicrons and VLDL) and cholesterol transport (associated with LDL and HDL).

LDL (LOW-DENSITY LIPOPROTEIN)

Although both LDL and HDL are primarily cholesterol particles, the majority of the cholesterol measured in blood is associated with **LDL**. The normal role of LDL is to deliver cholesterol to tissues for biosynthesis. However, cholesterol also plays an important role in cell membranes. In addition, bile acids and salts are made from cholesterol in the liver, and many other tissues require cholesterol for steroid hormone synthesis (steroidogenesis).

HDL (HIGH-DENSITY LIPOPROTEIN)

HDL is synthesized in the liver and intestines and released as dense, protein-rich particles into the blood. HDL contains apolipoproteins used for cholesterol recovery—that is, the cleaning up of excess cholesterol from blood vessels for excretion. HDL also delivers some cholesterol to steroidogenic tissues and transfers necessary apolipoproteins to some of the other lipoproteins.

Hyperlipoproteinemia

Fredrickson Classification

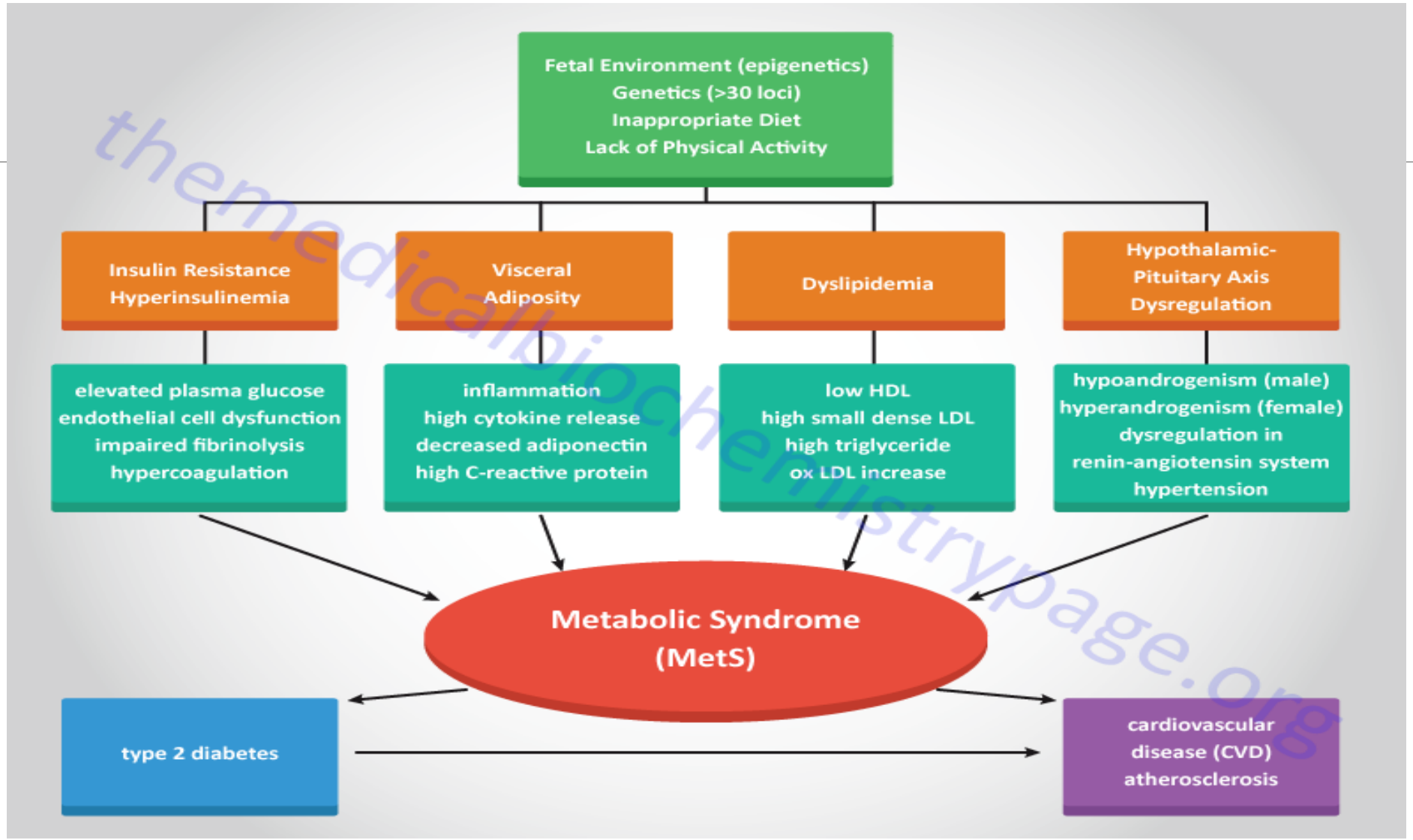
Type	Synonyms	Lipoprotein Elevation
I (rare)	''Primary hyperlipoproteinemia'', or ''Familial hyperchylomicronemia''	Chylomicrons
IIa	''Polygenic or Familial hypercholesterolemia''	LDL
IIb	''Combined hyperlipidemia''	LDL+VLDL
III (rare)	''Familial dysbetalipoproteinemia''	Chylomicrons+ IDL
IV	''Familial hyperlipemia''	VLDL
V (rare)	''Endogenous hypertriglyceridemia''	VLDL+ Chylomicrons

Hereditary Causes of Hyperlipidemia Caused by Known Single Gene Mutations

- Familial Hypercholesterolemia:
  - Occurs in 1 in 500 individuals/ AD.
  - Mutation in LDL receptor, resulting in elevated levels of LDL at birth and throughout life.
  - High risk for atherosclerosis, tendon xanthomas and xanthelasmas of eyes, CHD
- Familial Combined Hyperlipidemia:
  - Autosomal dominant.
  - Increased secretions of LDL& VLDLs.
  - High risk for atherosclerosis, no xanthomas.
- Dysbetalipoproteinemia:
  - Affects 1 in 10,000
  - A binding-defective form of apoE (which usually plays important role in catabolism of chylomicron and VLDL).
  - Increased risk for atherosclerosis, palmar xanthomas, CHD, PVD.

Secondary Causes of Hyperlipidemia

- Hypothyroidism (high LDL)
- NS (high LDL)
- Cholestasis (high LDL)
- Obesity (high TG)
- DM type 2 (high TG & chylomicrons)
- Pregnancy (high TG)
- Sepsis (high TG)
- Stress
- Reduced HDL: smoking, DM2, obesity, malnutrition, B- blockers
- Acute hepatitis (high TG)
- Drugs (thiazide, steroids, B- blockers, cyclosporine, protease inhibitors).
- MM, lymphoma (high TG)
- Glycogen storage disease (high TG).
- Alcohol, interferon, estrogen, thiazide, steroid (high TG).
- Acromegaly, renal failure (high TG).



http://themedicalbiochemistrypage.org/the-metabolic-syndrome-mets/

Glycerol Oxidation

95% of the energy in a fatty acid is derived from the oxidation of acetyl-CoA

5% from glycerol

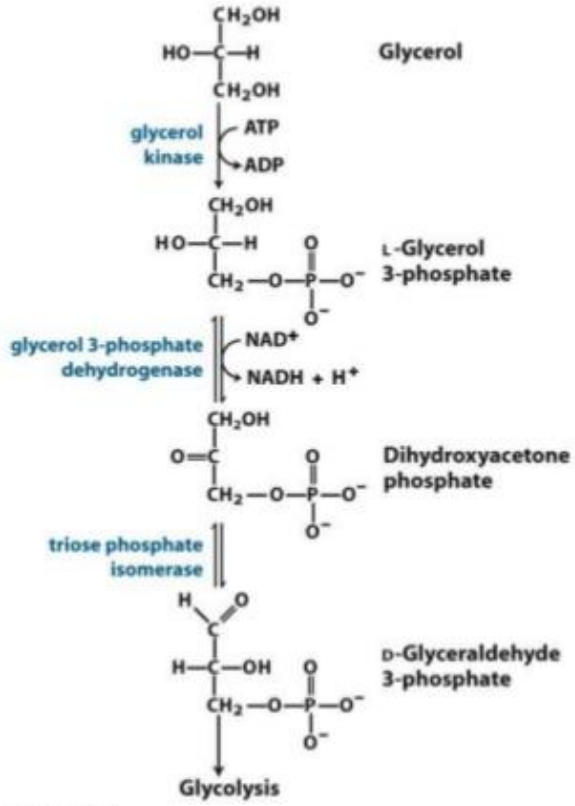


Figure 17-4
Lehninger Principles of Biochemistry, Fifth Edition

https://www.slideshare.net/obanbrahma/beta-oxidation-protein-catabolism-57804497

ATP Generation from Glycerol Oxidation

- glycerol – glycerol 3-phosphate - 1 ATP
- glycerol 3-phosphate - dihydroxyaceton phosphate 2.5ATP (1 NADH)
- glyceraldehyde 3-phosphate – pyruvate
- 4,5 ATP (1NADH + 2 ATP)
- pyruvate – acetyl CoA 2.5 ATP (1 NADH)
- acetyl CoA in Krebs cycle
  10 ATP (3NADH + 1 FADH2 + 1GTP)
- **Total** **19,5-1 = 18,5 ATP**

Energy balance of glycerol

Total ATP from glycerol:	
Glycerol to glycerol 3 phosphate	-1 ATP
Glycerol 3 phosphate to dioxyacetone -3-phosphate	+1 NADH = 3 ATP
Glycolysis (glyceraldehyde 3 phosphate to pyruvate)	+1 NADH + 2 ATP = 5 ATP
Pyruvate decarboxylation	+1 NADH = 3 ATP
Krebs cycle	+3 NADH = 9 ATP, 1FADH= 2 ATP, 1 GTP = 1 ATP

Total: 23 ATP – 1 ATP = 22 ATP

β oxidation of fatty acids

The **β oxidation** of long-chain fatty acids to acetyl-CoA is a central energy-yielding pathway. The electrons removed during fatty acid oxidation pass through the mitochondrial respiratory chain, driving ATP synthesis, and the acetyl-CoA

produced from the fatty acids may be completely oxidized to CO_2 via the citric acid cycle, resulting in further energy conservation. **β oxidation**, by which fatty acids are converted into acetyl-CoA.

The enzymes of fatty acid oxidation are located in the mitochondrial matrix.

The free fatty acids that enter the cytosol from the blood cannot pass directly through the mitochondrial membranes, but must first undergo a series of three enzymatic reactions. The first is catalyzed by a family of isozymes present in the outer mitochondrial membrane, acyl-CoA synthetases, which promote the general reaction:

Fatty acid + CoA + ATP fatty acyl-CoA + AMP + PPi

acyl-CoA

Fatty acyl-CoA esters formed in the outer mitochondrial membrane do not cross the inner mitochondrial membrane intact. Instead, the fatty acyl group is transiently attached to the hydroxyl group of **carnitine** and the fatty acyl-carnitine is carried across the inner mitochondrial membrane by a specific transporter In this second enzymatic reaction required for fatty acid movement into mitochondria, **carnitine acyltransferase I**, present on the outer face of the inner membrane, catalyzes transesterification of the fatty acyl group from coenzyme A to carnitine. The fatty acyl-carnitine ester crosses the inner mitochondrial membrane into the matrix by facilitated diffusion through the **acyl-carnitine/carnitine transporter**.

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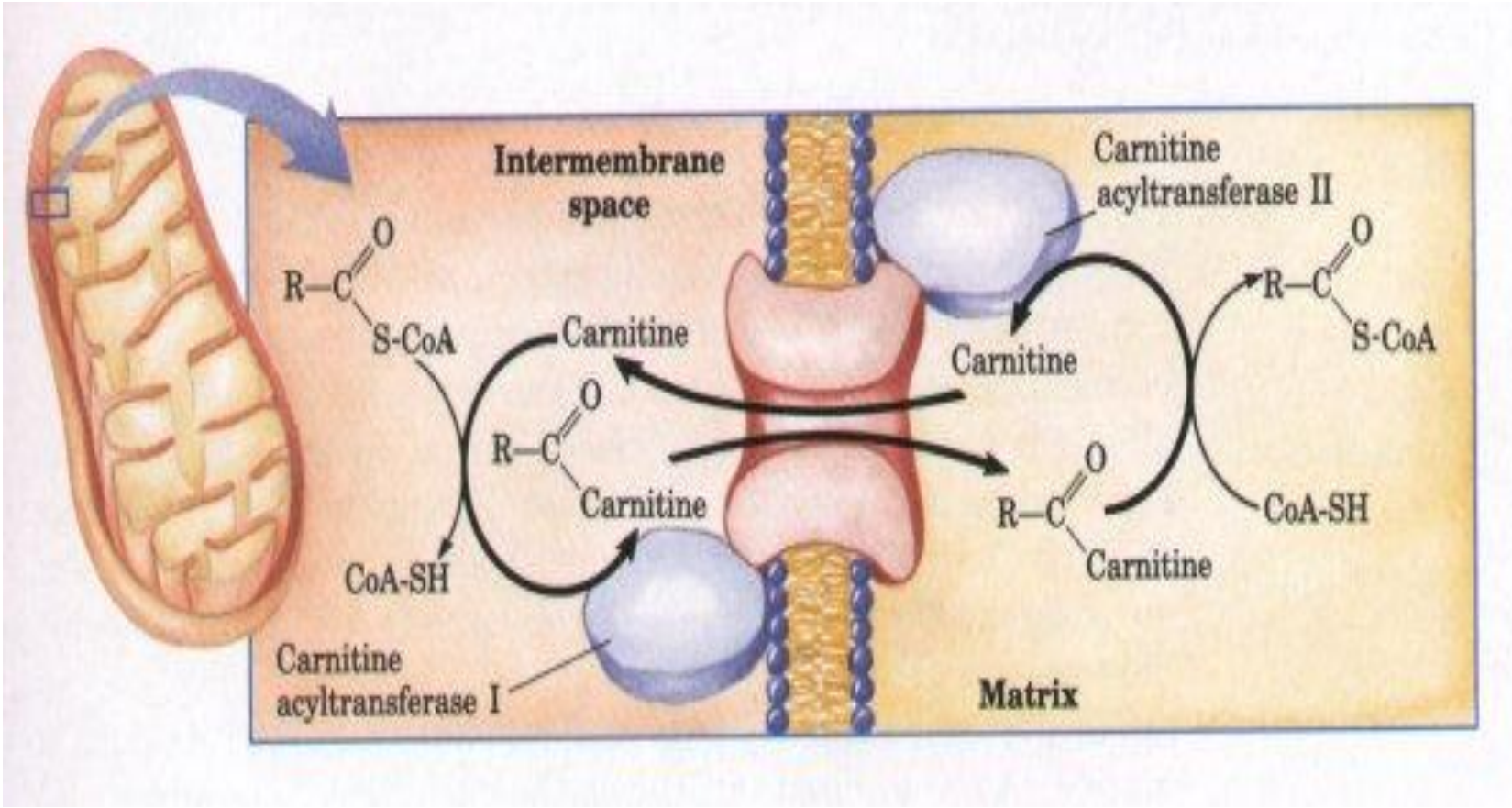
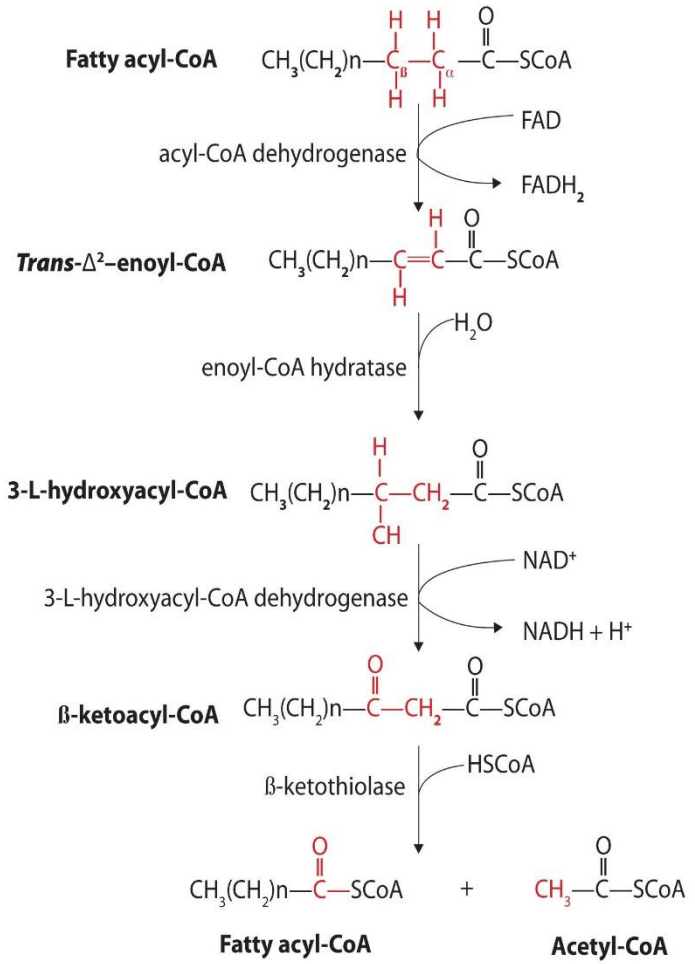


http://www.bioinfo.org.cn/book/biochemistry/chapt16/bio1.htm

Steps of β-oxidation



https://saylordotorg.github.io/text_the-basics-of-general-organic-and-biological-chemistry/s23-06-stage-ii-of-lipid-catabolism.html

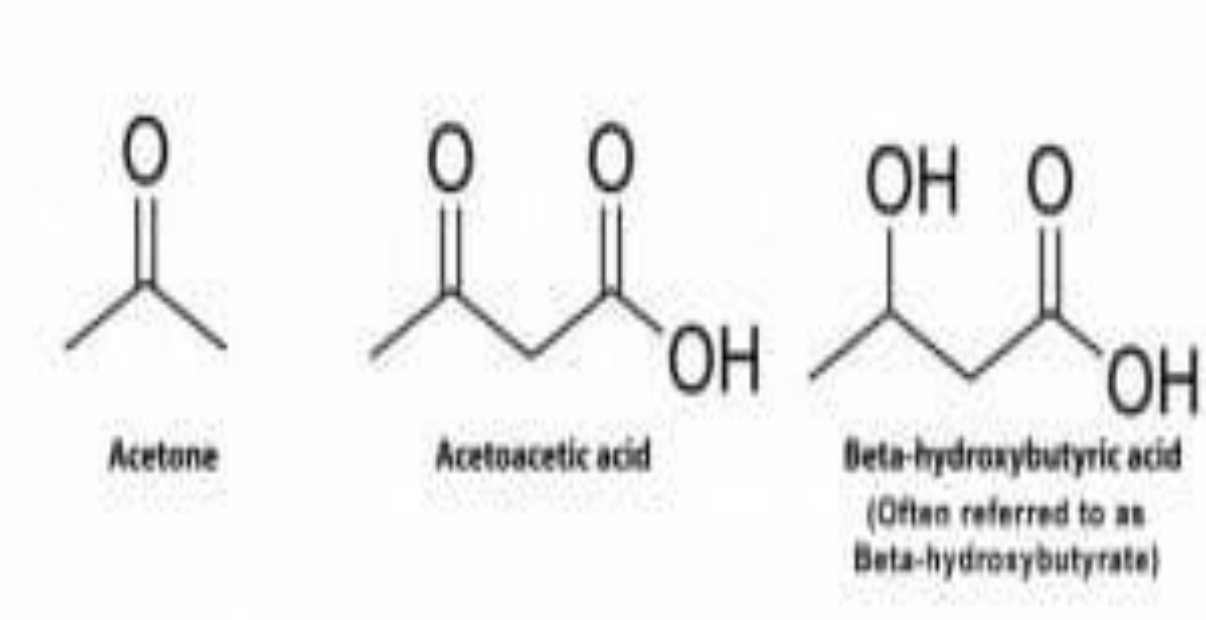
Energy Yield from β-Oxidation

- Yield of ATP per mole of stearic acid (C_{18}).

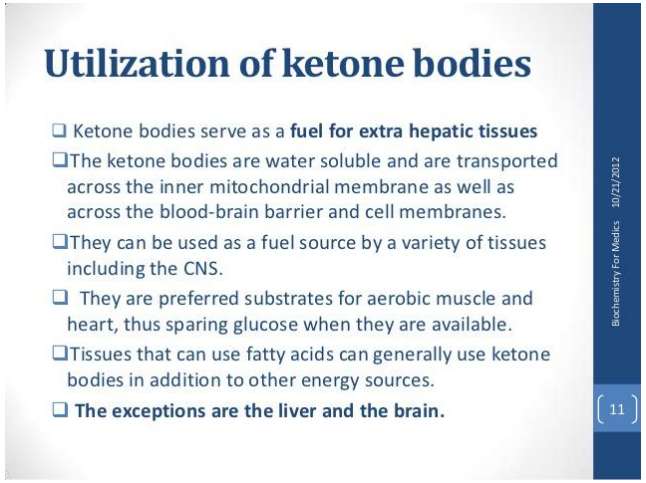
Step	Chemical Step	Happens	ATP
1	Activation (stearic acid -> stearyl CoA)	Once	-2
2	Oxidation (acyl CoA -> trans-enoyl CoA) produces $FADH_2$	8 times	16
4	Oxidation (hydroxy-acyl CoA to ketoacyl CoA) produces $NADH + H^+$	8 times	24
	Oxidation of acetyl CoA by the common metabolic pathway, etc.	9 times	108
		TOTAL	146

https://www.slideshare.net/obanbrahma/beta-oxidation-protein-catabolism-57804497

Ketone bodies



Ketone bodies, or simply *ketones* are substances produced by the liver from acetyl-CoA. There are three ketone bodies: *acetoacetate*, *beta-hydroxybutyrate*, and *acetone*. These compounds are used to provide energy to the cells of the body. There are important source of energy for cardiac muscle and neurons (when glucose is low or absent in the diet).
Concentration of ketone bodies increased in times of fasting and starvation, insulin dependent diabetes mellitus, intake of high in lipids and low in carbohydrates.



https://www.slideshare.net/namarta28/ketosis

Ketonemia

- **Ketonemia - increased concentration of ketone bodies in blood**
- **It is due to increased production of ketone bodies** by the liver rather than to a deficiency in their utilization by extra hepatic tissues.
- The production of ketone bodies occurs at a relatively low rate during normal feeding and under conditions of normal physiological status.
- Normal physiological responses to carbohydrate shortages cause the liver to increase the production of ketone bodies from the acetyl-CoA generated from fatty acid oxidation.

Causes of Ketosis

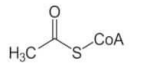
- ❑ Uncontrolled diabetes mellitus
- ❑ Starvation
- ❑ Chronic alcoholism
- ❑ Von- Gierke's disease
- ❑ Heavy exercise
- ❑ Low carbohydrate diet- For weight loss
- ❑ Glycogen storage disease type 6(Due to phosphorylase kinase deficiency)
- ❑ Pyruvate carboxylase deficiency

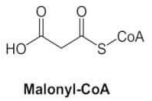
https://www.slideshare.net/namarta28/ketosis

Enzymes:
1.Fatty acid synthase (6 enzymes and 1 Acyl carrier protein molecule)
2.Acetyl-CoA carboxylase (rate-limiting enzyme)

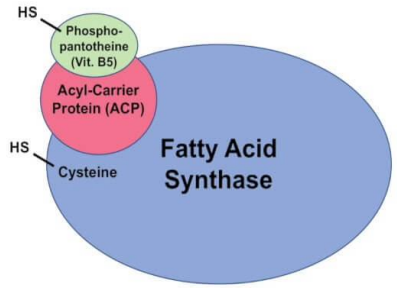
Starting material:
1.For palmitate synthesis: Acetyl-CoA
2.For odd number carbon long chain fatty acid synthesis: Propionyl-CoA
2 Carbon donor: Malonyl-CoA (donates 2 C and 1 C is thrown out as CO2)

Site: Cytosol
Citrate shuttle transfers acetyl-CoA from mitochondria to cytosol.
•OAA + **Acetyl-CoA** = Citrate (Mitochondria)
•Citrate (Mitochondria) = Citrate (Cytosol)
•Citrate = OAA + **Acetyl-CoA** (Cytosol)





Malonyl-CoA



Formation of **Malonyl-CoA** (3C) from Acetyl-CoA (2C) by **Acetyl-CoA carboxylase**.
•**1 ATP** used
•Requires **Biotin and Bicarbonate** (source of CO2)
•**Stimulated by:** Insulin, Citrate, ChREBP (induced by high carbohydrate diet/caloric intake)
•**Inhibited by:** Glucagon, Epinephrine (AMP dependent kinase), Palmitoyl-CoA
Malonyl-CoA inhibits carnitine acyltransferase to prevent fatty acids from being taken into the mitochondrial matrix to be beta oxidized at times when fatty acids are being synthesized, thus preventing a futile cycle.

It is a multimeric complex, but the 2 important domains are:
1. Ketoacyl synthase (KAS) end:
•has **Cysteine**-SH active site
•accepts **Acetyl-CoA and Acyl chain extended by 2 C on each cycle**
2. Acyl carrier protein (ACP) end:
•has **Pantothenic**-SH active site
•accepts **Malonyl-CoA**

https://epomedicine.com/wp-content/uploads/2018/07/FA-synthase.jpg

NADPH donors:
1.HMP shunt pathway
2.Isocitrate dehydrogenase
3.Malic enzyme
Mnemonic: HIM donates NADPH.

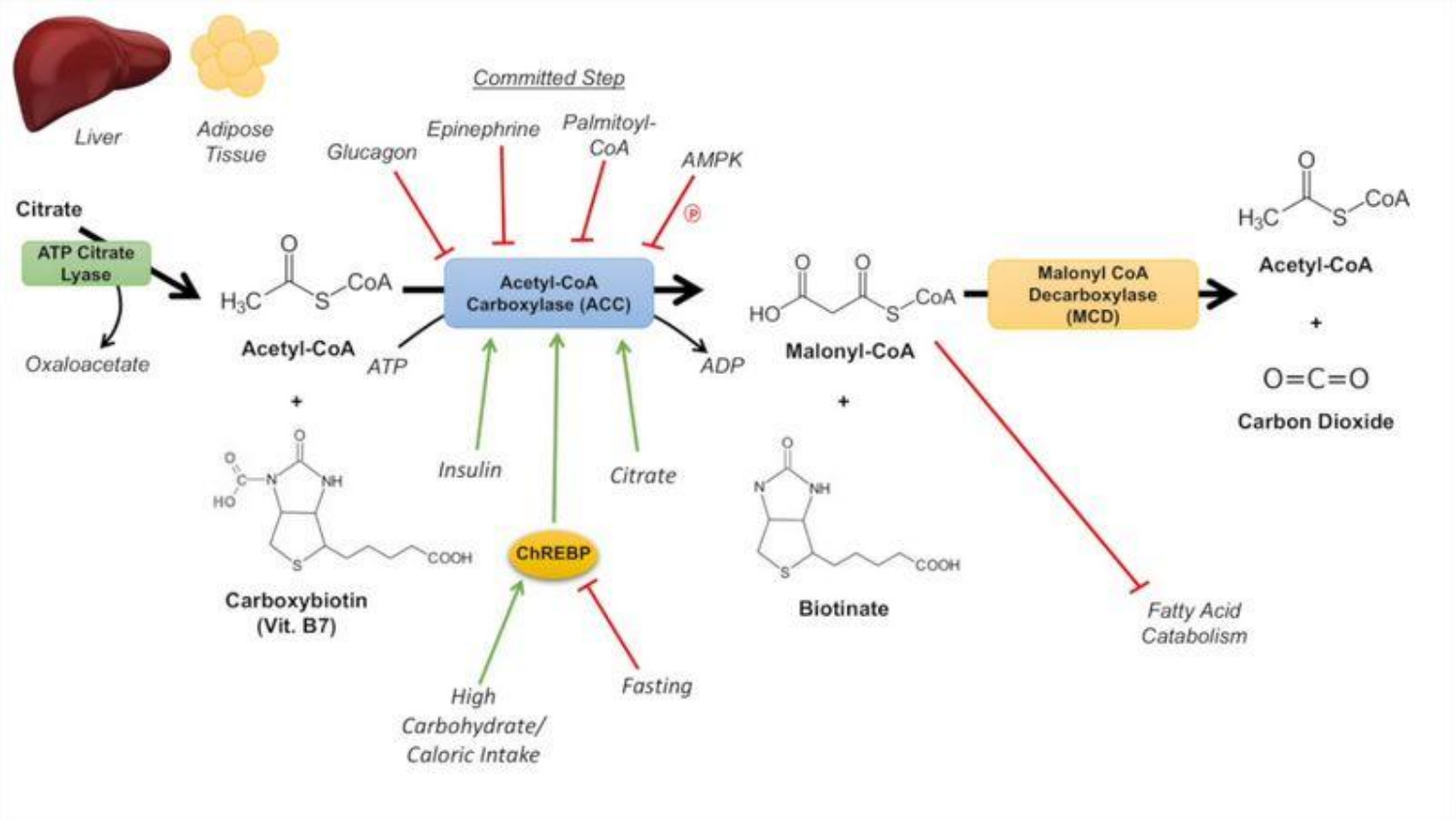
1. Loading of precursors to KAS and ACP end of FA synthase.
2. Condensation: Addition of 2 C atoms derived from Malonyl-CoA (Malonyl-CoA at ACP end takes precursor at KAS end and KAS end is free)
3. Reduction: **1 NADPH used**
4. Dehydration
5. Reduction: **1 NADPH used**
6. Acyl chain extended by 2 C atom (donated by Malonyl-CoA) is transferred to KAS end and ACP end is free to receive Malonyl-C0A.

Precursor loaded on KAS end is transferred to ACP end with Malonyl-CoA (KAS end is free).
Acyl chain elongated with 2 C atom is transferred from ACP end to KAS end (ACP end is free).
Malonyl-CoA is loaded on ACP end.
Cycle repeats.

7. **Cycle of precursor loading, condensation, reduction, dehydration and reduction** (Chain elongation) occurs unless **16 C Palmitate** is formed which is released from FA synthase enzyme complex by **Thioesterase**.

Humans make palmitic acid (16:0) as stored fat (only de novo fat possible).
•End-product: Palmitate (C16:0)
•Total of 7 cycles:
- •Starts with 2 carbon acetyl-CoA
- •Malonyl-CoA acts as a 2 carbon donor
- •7 Malonyl-CoA utilized (14 carbons)
- •2 NADPH (2 reduction reactions) X 7 cycles = 14 NADPH utilized
- •1 ATP (acetyl-CoA carboxylase reaction) X 7 cycles = 7 ATP utilized (+1 ATP to transport acetyl-CoA from mitochondria to cytosol)
- •1 CO_2 (released when malonyl-CoA donates 2 C acyl chain) X 7 cycles = 7 CO_2 released
- •1 Acetyl-CoA (to make malonyl-CoA) X 7 cylces + 1 Acetyl-CoA (precursor in KAS end) = 8 Acetyl-CoA utilized

Fatty acids synthesis



https://epomedicine.com/wp-content/uploads/2018/07/FA-synthase.jpg

Complex lipids

2. Complex lipids:

Esters of fatty acids with alcohols and molecules with other groups.

Phospholipids:

Lipids containing:

- Fatty acids
- Alcohol
- Phosphoric acid residue.

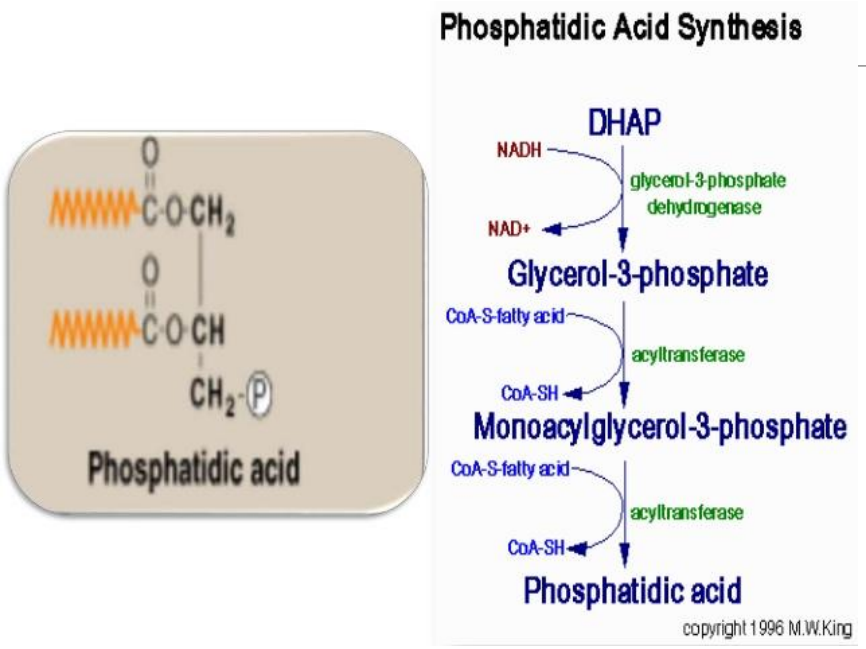
Glycolipids (glycosphingolipids):

Lipids containing:

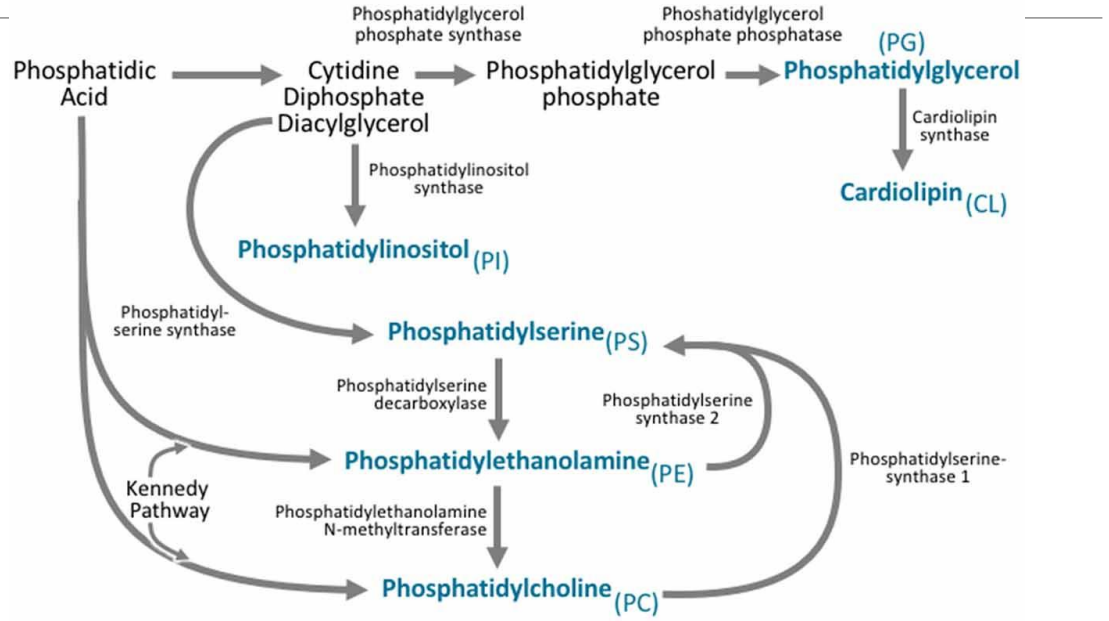
- Fatty acid
- Sphingosine
- Carbohydrate.

https://www.slideshare.net/Dr-HAMDAN/simple-lipids

Phospholipids synthesis



https://www.slideshare.net/YESANNA/synthesis-of-phospholipids



https://www.frontiersin.org/articles/10.3389/fnmol.2018.00010/full

Lipotropic factors

Lipotropic Factors

- These are the substances required for the normal mobilisation of fat from liver.
- The deficiency of these factors may result in fatty liver.
- Important lipotropic factors:
- Choline, Betaine, Methionine & Inositol.
- Folic acid, vitamin B_{12}, glycine & serine also serve as lipotropic factors.

https://www.slideshare.net/YESANNA/fatty-liver-43649058

Lipotropic factors

- Substances which facilitate mobilisation of fat and prevent accumulation of fat in liver are called LIPO TROPIC FACTORS / AGENTS / LIPOTROPINS

Lipotropic factors

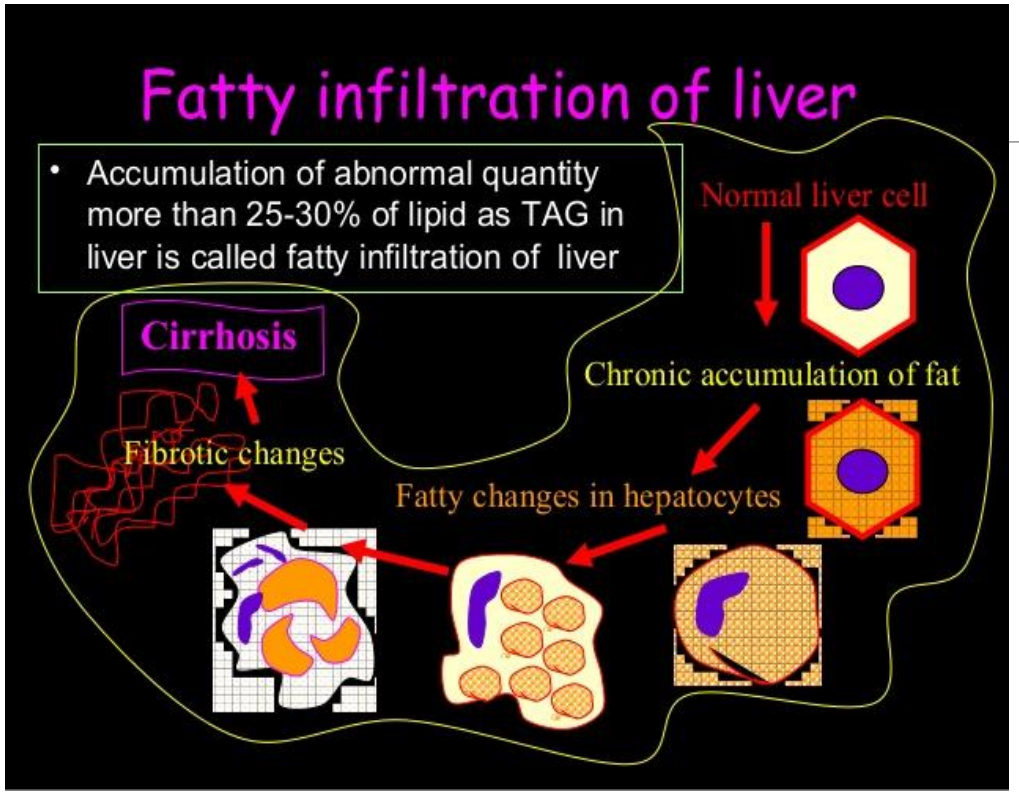
- **Choline** – ↓ oxidation of FA ↓phospholipid synthesis,
  – Impair lipoprotein syntheisis
- **Betain**-- ↓ choline synthesis,
  methyl gr. Donor for choline
- **Methionine**-- ↓ choline synthesis
- **Essential Fatty acids**– required for VLDL and lipoprotein synthesis
- **Inositol** -- ↓phospholipid synthesis

Lipotropic factors

- **Casein**
- **Heparin**
- **Vit. E, Selenium, Pantothenic acid**
- **ω3 Fatty acids** have protective role

- Lipotropic factors prevents fatty liver but cannot reverse the condition

https://pt.slideshare.net/DJ4SDM/fatty-liver-and-lipotropic-factors/4?smtNoRedir=1



https://pt.slideshare.net/DJ4SDM/fatty-liver-and-lipotropic-factors/4?smtNoRedir=1

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Chylomicrons%20are%20the%20least%20dense,HDL%20(high-density)

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