



Biochemistry

carbohydrates
proteins
isomers
ketone
starch
lipid
protein
amine

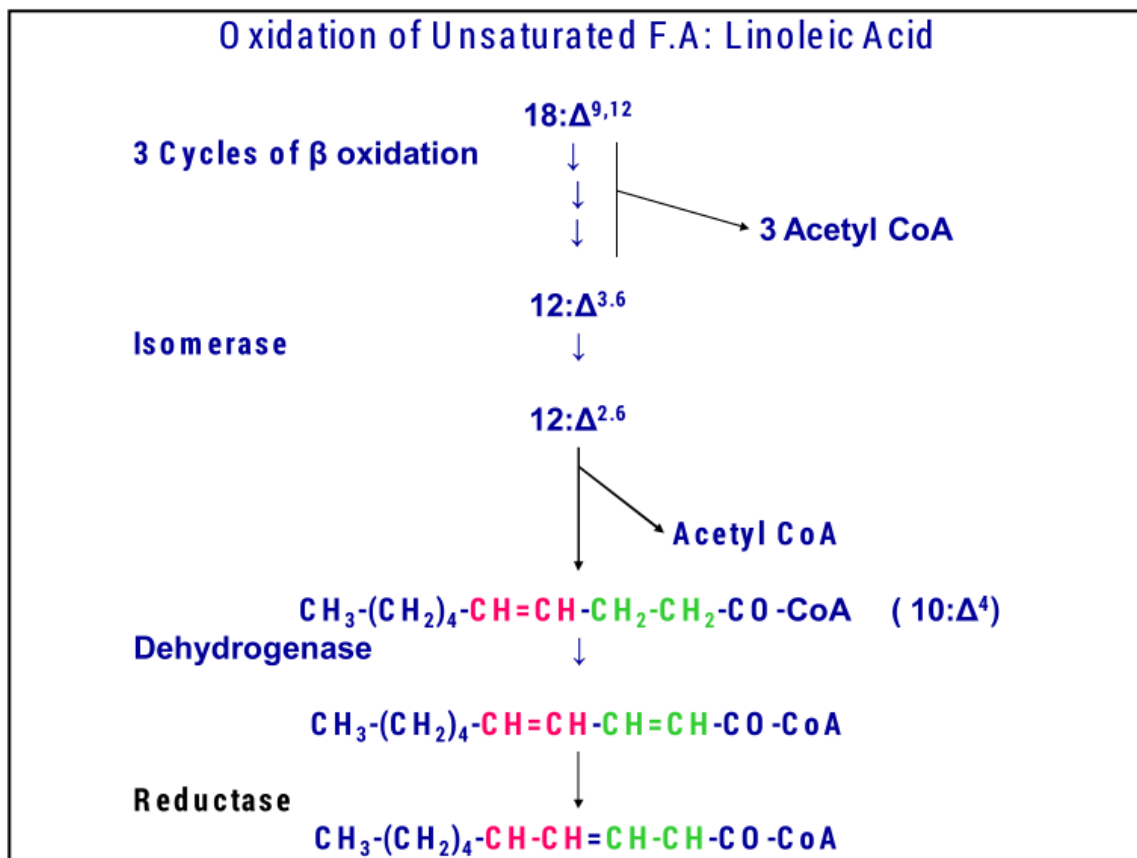
☒ Sheet

☐ Slides

Subject :	Fatty acid oxidation
Done by :	Ammar & Basheq
Corrected by :	Ola Al_juneidi
Number :	21

In the last lecture we talked about oxidation of unsaturated fatty acid which will be oxidized in the same manner as the saturated fatty acid except we may have double bond that isn't found in the proper place (the proper place is on C2 between C2 and C3) "remember in the oxidation we introduce a double bond on C2 in the first step ". The double bond may be on C3, so we need to shift this double bond to C2 by **Isomerase** as in the oleic acid.

In the case of linoleic acid (2 double bonds):



As we can see that linoleic acid is 18 carbons fatty acid with two double bonds on carbons 9 and 12.

- after 3 rounds of B-oxidation of this fatty acid, the resulting fatty acid will be 12 carbons fatty acid with two double bonds on C3 and C6 (since we removed the first 6 carbons in the form of 3 acetyl CoA) "we also get 3 NADH and 3 FADH₂."
- Now we have the double bond on C3 (inappropriate place), so we shift it to C2 using **isomerase**, so we can continue with an extra cycle to form another Acetyl CoA.

الدكتور ما حكاها "So now we have 4 cycles with (4 Acetyl CoA, 4 NADH, 3 FADH₂)" بس اتأكدوا منها. (we didn't need another FADH₂ because we already have a double bond)

- The resulting fatty acid is 10 carbons fatty acid with 1 double bond on C4 (in an appropriate place) so we produce another double bond between carbon 2 and 3 by **acyl CoA Dehydrogenase**, after that we will have 2 double bonds (at C2 and C4).

- This can't continue as usual, so **Reductase** will reduce the two double bonds (on c2 and c4) into one double bond that is on C3 (again , inappropriate place @_@) .

- the enzyme **isomerase** now will shift the double bond to C2 and we will continue another cycle and we will produce another Acetyl CoA .

- after this we will have 8 carbons saturated fatty acid that will undergoes 3 rounds of B-oxidation.

" the net 😊 : 9 Acetyl coA , 9 NADH , 7 FADH₂ "

الدكتور ما حكى عن 7 FADH₂ because we have 2 double bonds in the linoleic acid المحصلة عشان هيك اتأكدوا منهم

Note: Don't bother to calculate how much energy we get from the oxidation of unsaturated fatty acids (you just have to know the reactions, the enzymes and when they are needed). BUT you MUST know how to calculate the amount of energy produced (in the form of ATP or FADH₂ and NADH) from the oxidation of saturated fatty acids.

******So fatty acids with double bonds are oxidized as usual, and then we may require additional enzymes: **Reductase** and **isomerase** or **isomerase** alone according to the location of the double bonds.

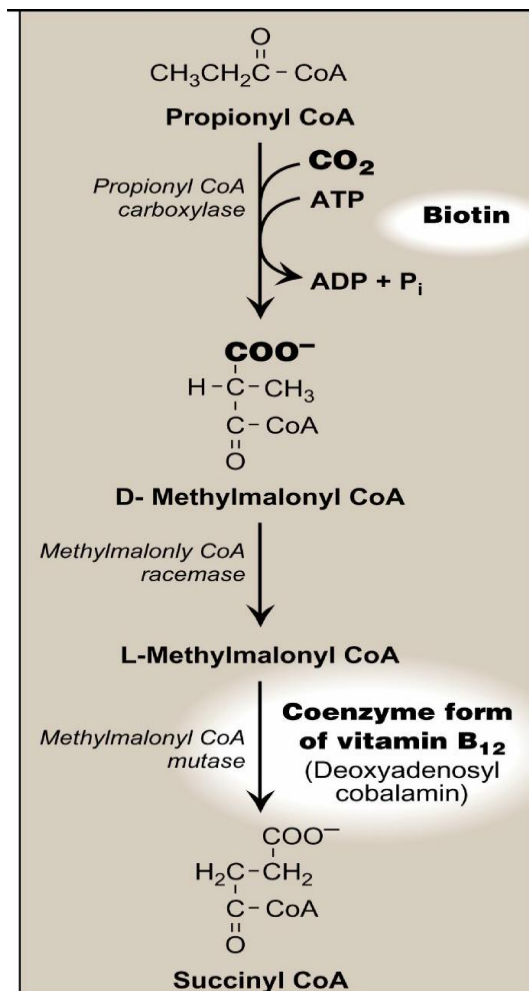
Oxidation of fatty acids with odd numbers :

These fatty acids aren't major in our bodies / food. Only 10% of animal fat may contain odd number fatty acids.

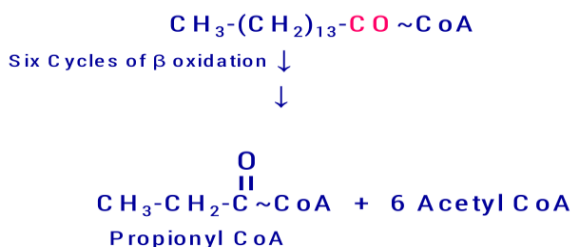
In this case we have a fatty acid with 15 carbons:

- Six rounds of β oxidation will proceed as usual; we remove two carbons each round, ending up with 3-carbons fatty acid (propionic acid / propionyl CoA).

[Keep in mind: Propionyl CoA can come from other pathway (we'll take it later)]



Oxidation of FA with odd number of carbons



▪ Propionyl CoA won't undergo oxidation as usual. However, it undergoes *carboxylation* (addition of CO_2 to the middle carbon) by the enzyme **propionyl CoA carboxylase** (it requires biotin as a co-enzyme to carry the carboxyl group).

☆ notice that we added the CO_2 to the second carbon.

▪ This reaction needs energy (ATP) because we added CO_2 to propionyl CoA.

Remember:

Decarboxylation reactions are EXERGONIC
 Carboxylation reactions are ENDERGONIC

▪ Now we have a 3-carbons fatty acid with 2 carboxyl carbons and an extra carbon (methyl) on carbon 2 [methylmalonyl CoA]

▪ this 3-carbons fatty acid is dicarboxylic acid just like the succinic acid (from the TCA cycle) but the succinic acid is 4-carbons fatty acid , and the malonic acid is 3-carbons fatty acid with methyl group on the middle carbon .

Now we switch the “D” form of Methylmalonyl CoA to the “L”.

▪ now the carboxylic carbon that we added before on carbon 2 will move to carbon 3 (that is the methyl in the methylmalonyl CoA) by **methylmalonyl CoA MUTASE** , and this will produce succinyl CoA which is an intermediate in TCA cycle.

00:00-13:30

☆ this reaction requires vitamin B12(cobalamin) . One of the two reactions in the body that requires vitamin B12. (Keep that in mind when we get to the other reaction)

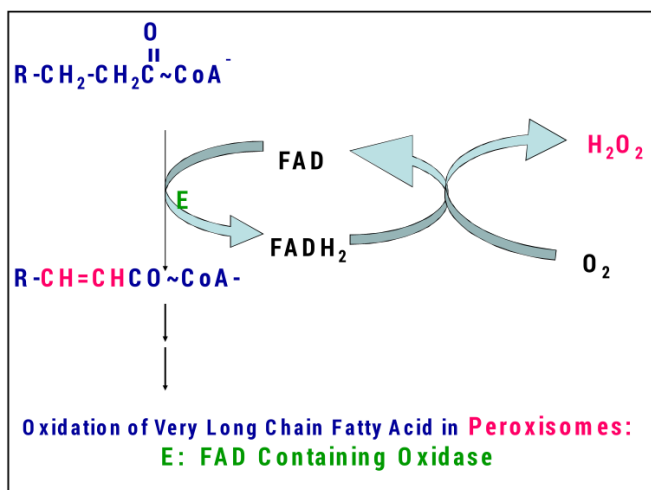
☆ if there is vitamin B12 deficiency (common), methylmalonyl CoA will accumulate >> methylmalonic acid will appear in the urine methylmalonyluria.

Nowadays you can measure the level of vitamin B12 in the plasma; we take a blood sample and give it to the laboratory. But this technique was not available like 30 years ago. So how did we measure it back then? We used to measure the levels of Methylmalonyl CoA, if it's elevated then we have a vitamin B12 deficiency.

▪ Succinyl CoA is an intermediate in the TCA cycle which will produce oxaloacetate at the end of the cycle and the oxaloacetate will produce glucose in the gluconeogenesis pathway. So, this is the only part of fatty acid that can be converted to glucose.

☆ Little amounts of fatty acid will be converted that are the last 3 carbons of the odd carbon fatty acid ONLY. Insignificant.

Oxidation of very long chain fatty acids > 20 C :



▪ these are oxidized in the peroxisome not in the mitochondria.

▪ the steps and the intermediates are almost the same to the saturated fatty acid metabolism but the enzyme that produce double bond between C2 and C3 is **FAD containing oxidase** which converts FAD to FADH₂.

Since the FADH₂ produced here can't go to the electron transport chain

(because there's no such thing in peroxisomes) and it can't go the mitochondria, it is reoxidized by the enzyme **oxidase** (NOT dehydrogenase).

*But what is the difference??

☆ **Oxidases** transfer the hydrogens to oxygen to produce H₂O or H₂O₂ while **Dehydrogenases** transfer the hydrogen to NAD or FAD or NADP.

☆ The “FAD” is a cofactor here not an electron acceptor. The enzyme is called “oxidase” because it uses Oxygen as an electron acceptor.

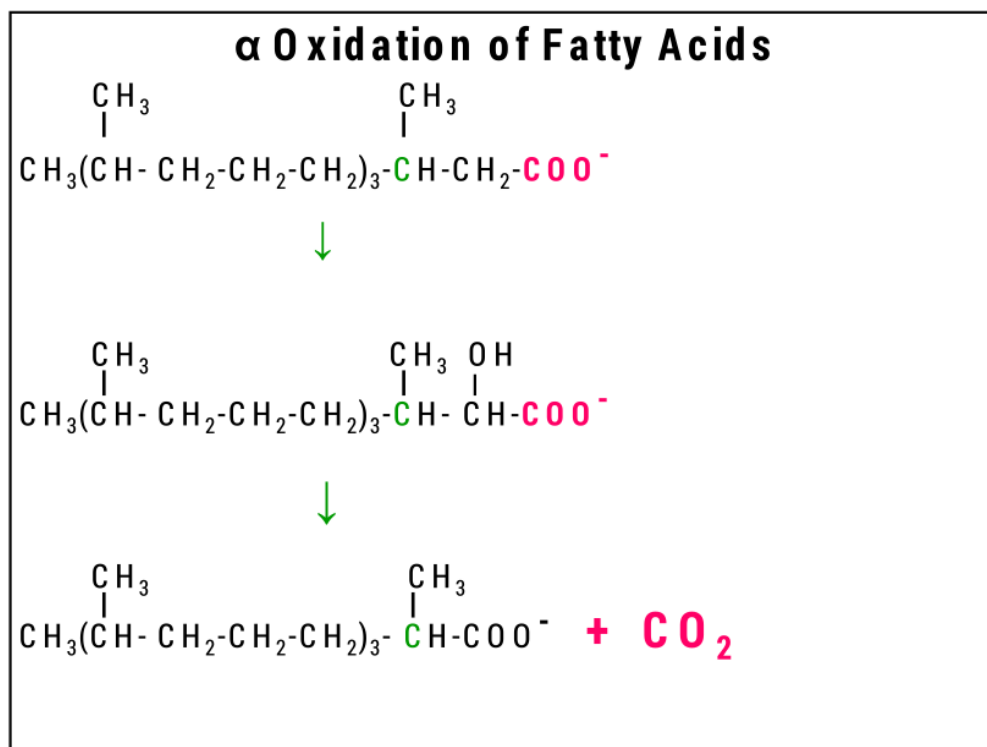
▪ O₂ will be reduced to H₂O₂ by reoxidizing the FADH₂ by certain enzymes.

☆ Remember: FADH₂ produced doesn't re-oxidize in the electron transport chain, because the reaction is not in the mitochondria.

▪ The reaction continues until the FA becomes suitable (medium or short chain) for β-oxidation so that it can be transported and oxidized in the mitochondria.

13:30-18:45

α – Oxidation of fatty acids:



▪ The oxidation occurs at carbon α (C2). We can't do beta oxidation because beta carbon has methyl group.

▪ Branched-chain fatty acids that have several methyl groups (every fourth carbon) (the first methyl group on carbon β).

▪ This kind of FAs associated with chlorophyll. (So it is present in our diets in vegetables but in minor amounts).

▪ α carbon is first hydroxylated (by oxidative hydroxylases).

- Then it is undergo *oxidative decarboxylation*; in which the carboxyl group leaves as CO₂, and the hydroxyl group is oxidized into carboxyl group.

After that the branch is now on the α-carbon and β-carbon (that was gamma carbon) becomes free. Now oxidation occurs as usual.

- It is a minor pathway but it has a clinical significant, in a rare genetic disease in which one of the enzymes is missing resulting in accumulation of these FAs in brain cells leading to neurological disorders. To avoid this, the patient should avoid food that contains branched FA (simply!).

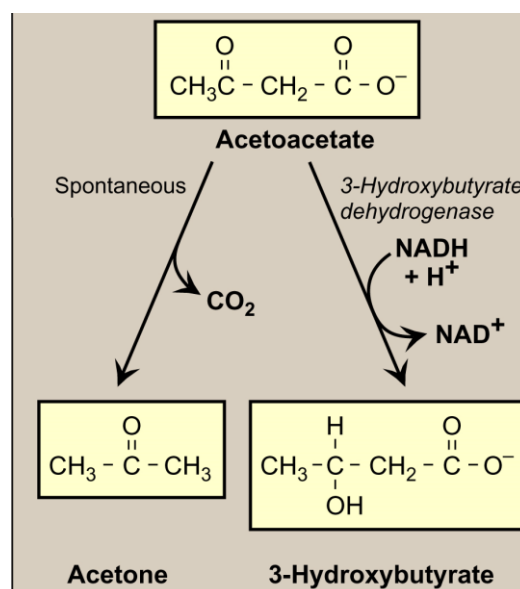
Ketone bodies :

As we know, the end product of β oxidation of FAs is acetyl CoA, which can be oxidized completely by TCA (Krebs's cycle) or it can be converted into ketone bodies

*What are ketone bodies?

They're 3 compounds related to β oxidation (and Acetyl CoA is their precursor):

- The first structure (acetoacetate) is a derivative of butyric acid (Ketobutyric acid), the left half of this compound (CH₃CO-) is acetic acid, and the right half is acetate, acetic group and acetic group linked together, so it's named **acetoacetate**.
- Acetoacetate may undergoes spontaneous decarboxylation (the carboxyl group to the right is lost as CO₂ without an enzyme) to give a ketone compound called **acetone**, which is the second ketone body.
- Acetoacetate can also be reduced by NADH (oxidizing it to NAD⁺) so the ketone group is converted to a hydroxyl group to give **3-Hydroxybutyrate**, which is considered the third ketone body although it is not actually a ketone.



These ketone bodies (Acetoacetate, Acetone, and 3-Hydroxybutyrate) are synthesized in the liver all the time at low rates. But during fasting and uncontrolled diabetes mellitus (Type1), they will be synthesized at high rates.

The pathway of their synthesis has 3 steps:

The first step is a condensation of 2 acetyl CoA molecules producing Acetoacetyl CoA, by an enzyme called **thiolase**.

Notice that this product is the last intermediate in β oxidation, the last step in β oxidation is the cleavage of Acetoacetyl CoA to 2 acetyl CoA. Therefore, high amounts of acetyl CoA whether it's coming from β oxidation or from any other source will increase the rate of this condensation reaction.

(Some amino acids can be directly degraded to Acetoacetyl CoA)

The second step involves the addition of one more Acetyl CoA to Acetoacetyl CoA, producing a product called **HMG CoA**. So named because if you look at its structure you'll notice that it is a 5-carbon chain with 2 carboxyl groups at each end, it's similar to glutaric acid which is a 5-carbon dicarboxylic acid.

So HMG CoA is derivative of glutaric acid, but it is modified by addition of methyl and hydroxyl groups at the third carbon, so the name becomes 3-hydroxy-3-methylglutaryl CoA (abbreviated HMG CoA.)

The enzyme responsible for this step is **HMG CoA synthase** (notice: not synthetase because there is no ATP involved in the reaction)

The third step is cleavage of HMG CoA by *HMG CoA lyase*, acetyl CoA is removed (**not the one added in step 2**) and what remains is acetoacetate.

*What is the net reaction of this pathway?

(NOTE: Any intermediate in the pathway is cancelled from the net reaction, meaning that any compound that is produced in one step and used or removed in another step is not counted in the net reaction, such as acetyl CoA (the third one) and HMG CoA which are intermediates.

So they're not mentioned, therefore the net reaction is:



From the net reaction you can tell the purpose of the pathway for these ketone bodies, there are three basic purposes:

- Production of CoA (regeneration)
- Production of Acetoacetate
- Removal of Acetyl CoA

For the liver, Acetyl CoA is the important product (Acetoacetate is a by-product), while acetoacetate is used in other tissues like muscles.

Palmitic acid as an example (16-carbon fatty acid) is oxidized by β oxidation to give 8 acetyl CoA, and these acetyl CoA groups are **completely degraded** to give 16 CO₂, 7 NADH, 7 FADH₂, and **8 coenzyme A molecules are regenerated and released to function again in β oxidation.** (NOTE: NADH and FADH₂ are used in ETC for energy production).

****Keep in mind that β oxidation requires [CoA, NAD⁺, FAD, And ATP for activation of FA] so we need continuous supply of them.**

*What is the fate of acetyl CoA?

--It will enter TCA cycle (combine with oxaloacetate producing citrate).

*Is oxaloacetate required in the same amount as acetyl CoA?

--NO, because every time oxaloacetate is used, it is regenerated by the cycle.

CoA is found in limited quantities. So we have to regenerate it so β -oxidation won't stop. Under normal conditions it is released from Acetyl CoA through the steps of TCA cycle.

28:00-39:00

During Fasting, gluconeogenesis occurs in the liver and oxaloacetate is consumed, (oxaloacetate is an intermediate in TCA cycle and is normally found in small "catalytic" constant concentration, it is constant because its rate of consumption is equal to production), so when all the oxaloacetate is consumed in gluconeogenesis TCA cycle is stopped and as result, CoA is locked (trapped) in Acetyl CoA.

B-oxidation also occurs during fasting, but requires CoA ... How can we regenerate CoA when TCA is not running?? By production of ketone bodies! So the aim of ketone bodies' production is to regenerate CoA that is essential for β oxidation to keep running and producing energy (in the form of NADH and FADH₂).

Notice that FAs in the case of fasting are not completely oxidized to CO₂, only beta oxidation occurs (citric acid cycle is not functioning that's why we don't get energy from acetyl CoA). Even though, fatty acid oxidation is the major source of energy during starving (*this is similar to the conditions of hypoxia, where anaerobic respiration is the major source of energy although it produces only small amounts of energy*)

Again, during prolonged fasting, glucagon activates hormone-sensitive lipase, which results in high amounts of FA reaching the liver. Ketone bodies production is increased due to:

- Increased concentration of substrates (FAs which are **partially** oxidized to produce Acetyl CoA)
- Absence of oxaloacetate >> TCA cycle turned off >> No CoA regeneration

In uncontrolled diabetic patients:

Blood glucose is very high (3 or 4 times higher than the normal glucose level) so the liver is expected to lower the production of glucose, yet the liver is actively making glucose although the blood glucose level is high.

This is because the liver does not sense the level of glucose in blood; it senses hormones; insulin and glucagon. **Insulin is low, glucagon is high, and they tell the liver to do gluconeogenesis.**

So the liver continues to do gluconeogenesis even though the blood glucose is high because it responds to insulin and glucagon but not the glucose level in blood. (Here the liver is acting as it senses a starvation).

Active gluconeogenesis in diabetes, active lipolysis, free fatty acids in the plasma are high, hepatic output of ketone bodies is high, and this all results in **ketoacidosis**.

Ketoacidosis is a drop of blood pH caused by the release of ketone bodies from the liver (remember that they are not used by the liver at all), this result is expected since two of the ketone bodies are acids.

ketoacidosis results in increase excretion of ketone bodies in urine (the level of acetoacetate and β -Hydroxybutyrate is very high in the plasma so they will be excreted in the urine), they are anions, negatively charged, so they are excreted as sodium salts, and loss of sodium ions results in loss of water. As a result **dehydration** occurs.

The **diabetic ketoacidosis (DKA)** is very common in uncontrolled diabetes, and it is an emergency case. DKA occurs mainly in type 1 diabetic patients (insulin dependent), that's why it is seen in children much more than in adults.

If it continues it leads to loss of consciousness and coma and if persists it may lead to death.

(NOTE: this is accompanied by the smell of acetone from the mouth, this

acetone is one of the ketone bodies that are produced and it exits because it is volatile)

We deal with such cases (coma) by the use of IV fluids (to compensate for dehydration) and the injection of glucose and insulin to restore normal functions, the improvement is dramatic and it only takes 5 to 6 hours.

The ketone bodies produced during prolonged fasting are released to the plasma and taken up by other tissues, like muscle tissue.

There Hydroxybutyrate can be oxidized to acetoacetate, then acetoacetate can be consumed but it requires CoA, it should be linked to CoA to produce Acetoacetyl CoA.

Succinyl CoA (an intermediate in TCA) is the donor of CoA, it provides CoA for acetoacetate to be converted into **Acetoacetyl CoA** which can be cleaved into 2 acetyl CoA to be used in the citric acid cycle in the muscles. (So muscles [especially the cardiac muscle] use these acetyl CoA as source of energy because the amount of oxaloacetate is adequate, *no gluconeogenesis in muscles*).

39:00-49:45

What happens for concentrations during starvation?

- Concentration of fatty acids in the second or third day increases almost 3 times, fatty acids in high concentrations in the circulation.
- Concentration the of ketone bodies increases 10-20 times, so they're highly available and can be used as a source of energy.
- Glucose is maintained at 80% by gluconeogenesis.

Normally, the brain is completely dependent on glucose; fatty acids are not a source of energy for the brain. However, during prolonged fasting the brain can use ketone bodies as a source of energy.

The usage of ketone bodies by the brain spares and preserves our body's proteins, which are otherwise were going to be degraded to provide glucose for the brain.

Fuel metabolism in starvation

	Amount formed or consumed in 24 hours (grams)	
Fuel exchanges and consumption	3rd day	40th day
Fuel use by the brain		
Glucose	100	40
Ketone bodies	50	100
All other use of glucose	50	40
Fuel mobilization		
Adipose-tissue lipolysis	180	180
Muscle-protein degradation	75	20
Fuel output of the liver		
Glucose	150	80
Ketone bodies	150	150

Note that the amount of ketone bodies used as source of energy is doubled, which contributes to the lowering of muscle protein degradation from 75 to 20, and this an advantage for survival