**The Synthesis and Utilization of Ketone bodies**

Ketone bodies are two molecules, acetoacetate and \( \beta \)-hydroxybutyrate. The term “ketone body” is historical: only acetoacetate is an actual ketone. Ketone bodies are synthesized in the liver from acetyl-CoA.

The brain is an important organ. It is metabolically active and metabolically privileged. The brain generally uses 60-70% of total body glucose requirements, and always requires some glucose for normal functioning. Under most conditions, glucose is essentially the sole energy source of the brain. The brain cannot use fatty acids, which cannot cross the blood-brain barrier. Because animals cannot synthesize significant amounts of glucose from fatty acids, as glucose availability decreases, the brain is forced to use either amino acids or ketone bodies for fuel. Individuals eating diets extremely high in fat and low in carbohydrates, or starving, or suffering from a severe lack of insulin (Type I diabetes mellitus) therefore increase the synthesis and utilization of ketone bodies.

Ketone body synthesis occurs normally under all conditions. However, the formation of ketone bodies increases dramatically during starvation. This seems to be due to a combination of factors. Prolonged low levels of insulin result in both increased fatty acid release from adipose tissue, and increased amounts of the enzymes required to synthesize and utilize ketone bodies. In addition, in the liver, increased demand for gluconeogenesis results in depletion of oxaloacetate, and therefore in decreased capacity for the TCA cycle. This causes a rise in the levels of acetyl-CoA, the substrate for ketone body production.

The first enzyme in the ketone body synthesis pathway is thiolase (the same enzyme that is responsible for the cleavage step in \( \beta \)-oxidation). In ketone body biosynthesis, thiolase catalyzes the condensation of two acetyl-CoA molecules to form acetoacetyl-CoA. The next enzyme, HMG-CoA synthase adds a third acetyl-
CoA molecule, to form \( \text{[\text{-}hydroxy-\text{-}methylglutaryl-CoA} \) (usually abbreviated HMG-CoA). HMG-CoA is an important biosynthetic intermediate; however, in the mitochondria, it is only used for ketone body synthesis. The third enzyme, HMG-CoA lyase, releases an acetyl-CoA from HMG-CoA to form acetoacetate.

The final enzyme in ketone body synthesis, \( \text{[\text{-}hydroxybutyrate dehydrogenase} \), reduces acetoacetate to \( \text{[\text{-}hydroxybutyrate} \). The \( \text{[\text{-}hydroxybutyrate dehydrogenase} \) reaction has two functions: 1) it stores energy equivalent to an NADH in the ketone body for export to the tissues, and 2) it produces a more stable molecule. Acetoacetate is a \( \text{-ketoacid} \), and like many such compounds may spontaneously decarboxylate. The product of the decarboxylation reaction, acetone, is a volatile waste product, and is largely excreted via the lungs. “Acetone-breath” has been used as a crude method of diagnosing individuals with untreated Type I diabetes mellitus.

Both \( \text{[\text{-}hydroxybutyrate} \) and acetoacetate are released into circulation. The ratio depends on the amount of NADH available in the liver mitochondria; if NADH concentration is high, the liver releases a higher proportion of \( \text{[\text{-}hydroxybutyrate} \).

Both \( \text{[\text{-}hydroxybutyrate} \) and acetoacetate are organic acids. These compounds are released in the protonated form, which means that their release tends to lower the pH of the blood. In normal individuals, other mechanisms compensate for the increased proton release. Individuals with untreated Type I diabetes mellitus often release ketone bodies in such large quantities that the normal pH-buffering mechanisms are overloaded; the reduced pH, in combination with a number of other metabolic abnormalities associated with lack of insulin results in diabetic ketoacidosis, a life-threatening acute disorder of Type I diabetes. In most cases, the increase in ketone body concentration in blood is due to increased synthesis in liver; in severe ketoacidosis, cells begin to lose ability to use ketone bodies also.

**Why are three enzymes required to synthesize acetoacetate?** An enzyme that cleaves the thioester bond of the thiolase product acetoacetyl-CoA would also produce acetoacetate, but such a thioesterase does not seem to exist. The reason for the multienzyme pathway is not really understood. However, the pathway that does exist is not especially wasteful; the third acetyl-CoA used merely acts catalytically. Because the cell needs to have HMG-CoA synthase for other purposes, the choice is in having HMG-CoA lyase, or our imaginary thioesterase. It is possible that having two mitochondrial enzymes (HMG-CoA synthase and HMG-CoA lyase) required for ketone body synthesis assists in controlling the pathway.

**Control of ketone body synthesis**

Several factors influence ketone body production. Ketone body production only occurs during conditions of high circulating free fatty acids. One possible fate for the fatty acids is ketone body production, while another possible fate is conversion to triacylglycerol. However, because the glycerol required for triacylglycerol synthesis is derived from glycolysis, when glycolytic and gluconeogenic substrates are limiting the liver will make primarily ketone bodies. Finally, the liver must have excess...
energy. High levels of circulating fatty acids provide the energy to allow the liver to divert excess acetyl-CoA to ketone body synthesis.

Prolonged low levels of insulin stimulate production of mitochondrial HMG-CoA synthase and of HMG-CoA lyase. These elevated enzyme levels, in combination with increased acetyl-CoA substrate availability, and lack of gluconeogenic substrates, favor ketone body production.

**Ketone body utilization**

Tissues that can use fatty acids can generally use ketone bodies in addition to other energy sources. The exceptions are the liver and the brain. The liver synthesizes ketone bodies, but has little $\beta$-ketoacyl-CoA transferase, and therefore little ability to convert acetoacetate into acetyl-CoA. The brain does not normally use fatty acids, which do not cross the blood-brain barrier; under ordinary circumstances, the brain uses glucose as its sole energy source.

The metabolic rate of the brain is essentially constant. While other tissues reduce their metabolic requirements during starvation, the brain is unable to do so. After a few days of fasting, the brain undergoes metabolic changes to adapt to the decreased availability of glucose. One major change is increased amounts of the enzymes necessary to metabolize ketone bodies.

The utilization of ketone bodies requires one enzyme not present in the ketone body biosynthetic pathway, $\beta$-ketoacyl-CoA transferase, converts acetoacetate to acetoacetyl-CoA. As mentioned above, lack of this enzyme in the liver prevents the futile cycle of synthesis and breakdown of acetoacetate.

![Diagram](image)

The $\beta$-ketoacyl-CoA transferase uses succinyl-CoA as the CoA donor, forming succinate and acetoacetyl-CoA. This reaction bypasses the succinyl-CoA synthetase step of the TCA cycle, although it does not alter the amount of carbon in the cycle. This implies that the TCA cycle must be running to allow ketone body utilization; a fact which is necessarily true, because the TCA cycle is necessary to allow generation of energy from acetyl-CoA.
The other enzymes of the ketone body utilization pathway, \(-\)-hydroxybutyrate dehydrogenase and thiolase, are identical to the enzymes used for ketone body synthesis.

**Summary**
One possible fate for the fatty acids released as a result of triacylglycerol breakdown is conversion to ketone bodies. Ketone bodies, unlike fatty acids, cross the blood-brain barrier. Ketone bodies, unlike glucose, can be synthesized from acetyl-CoA. Ketone bodies can therefore provide energy to the brain when glucose availability is limited.

Ketone bodies are synthesized from acetyl-CoA in liver mitochondria by a short pathway beginning with thiolase. One control for the production of ketone bodies is the fact that high levels of acetyl-CoA favor the thiolase condensation reaction that forms acetoacetyl-CoA, rather than the thiolase cleavage reaction that produces additional acetyl-CoA.

Ketone bodies are converted back to acetyl-CoA using \(-\)-ketoacyl-CoA transferase. Liver mitochondria lack this enzyme; starvation causes the brain and some other tissues to increase the synthesis of \(-\)-ketoacyl-CoA transferase, and therefore to increase their ability to use these compounds for energy.