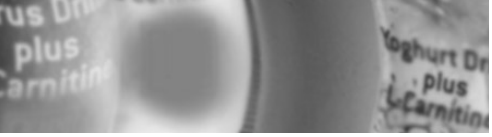

100 years of L-Carnitine. L-Carnipure® L-Carnitine and fatty acid oxidation

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Carnitine, or 3-hydroxy-4-N-trimethylammonium butyrate, is a naturally occurring compound which marks the 100th anniversary of its original discovery in skeletal muscle extracts this year (1,2). L-Carnitine, the biologically effective isomer of carnitine, plays a key role within several cellular energy producing pathways. By way of example, L-Carnitine is essential towards the transport of long-chain fatty acids across the inner-mitochondrial membrane towards their oxidative fate inside the mitochondrial matrix, is important towards the removal of toxic acyl-CoAs from the mitochondria by forming acylcarnitines and serves as a temporal acetyl group buffer during the oxidation of carbohydrates during periods of increased pathway flux.

WHAT IS β-OXIDATION?

Lipids are energy-rich substances that serve as a major source of fuel for the body's metabolic processes. Fats further play an important role as a constituent of cell membranes and are essential for the supply of fat-soluble vitamins and unsaturated fatty acids. Fats are obtained from food or formed in the body, mostly in the liver, and can be stored in fat cells for future use. On average, a man of 70 kg body weight is assumed to have stored 135,000 kcal of energy in form of fat in his adipose tissue. Fat is a poor thermal conductor and does not transfer heat very well. Thus the layer of fat beneath the skin acts as an insulator, helping to maintain the body temperature. In addition it serves as a shock absorber, supporting and cushioning the vital organs. From storage sites, the fats are mobilised and released as free fatty acids. These are transported in the plasma to the site of their oxidation.

The β-oxidation of long-chain fatty acids is central to the provision of energy for the organism and is of particular importance for the heart and skeletal muscle (3). During this process, activated long-chain fatty acids are broken down into 2-carbon acetyl-fragments. The acetyl groups subsequently combine with Coenzyme A (CoA) to form acetyl-CoA, which is metabolised in the Krebs' cycle and respiratory chain with the production of H_2O and CO_2. The resulting product of these processes is adenosine triphosphate (ATP) to realize the energy metabolism of the cell.

The efficiency with which glucose or fatty acids provide ATP differs somewhat, however. A single mole of palmitate (a 16-carbon fatty acid) catabolised through β-oxidation will provide 8 moles of acetyl-CoA in the Krebs' Cycle, whereas a single mole of glucose (6-carbon) provides only 2 moles of acetyl-CoA.

β-oxidation takes place inside the mitochondria, but long-chain fatty acids are unable to penetrate the inner mitochondrial membrane. Therefore there is a need for a carrier to get the long-chain fatty acids to the place of oxidation (4). This carrier is L-Carnitine.

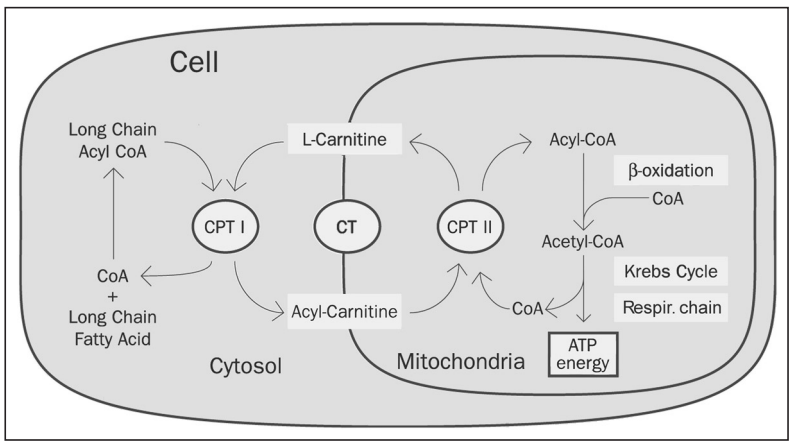
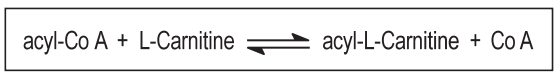


Figure 1 – Cell – How it works

"L-CARNITINE TURNS FAT INTO ENERGY"

In 1905, L-Carnitine was isolated for the first time from muscle tissue (2), and its structure was established in 1927 (5). The body can synthesize L-Carnitine from protein-bound Lysine and Methionine, but for the major part, however, the daily L-Carnitine requirement is met by food intake. Products of animal origin contain the highest amounts of dietary L-Carnitine, whereas plant products contain only traces of this substance. For biosynthesis, the body also requires sufficient supply with vitamins C, B3, B6 and iron. Whereas the steps of synthesis of the last precursor of L-Carnitine, 4-butyrobetaine, can be performed in practically all tissues of the body, the last step, which is a hydroxylation reaction, almost exclusively is done in the liver. The L-Carnitine-rich skeletal muscles are not able to synthesize L-Carnitine themselves.

The inner mitochondrial membrane is not permeable to activated fatty acids from the cytosol. Therefore, long-chain fatty acids have to be transported into the mitochondrial matrix via a regulated carrier system for their β-oxidation. The fatty acid transfer system consists of three enzymatic steps: Carnitine palmitoyltransferase 1 (CPT-1), Carnitine translocase and Carnitine palmitoyltransferase 2 (CPT-2). Thus L-Carnitine is involved in a transfer mechanism, where long-chain fatty acid-CoA-esters are transformed into the corresponding Carnitine esters and shuttled into the matrix space of the mitochondria (Figure 1).



Scheme 1

A well-known function of L-Carnitine in metabolism is as a substrate for a series of enzymes, Carnitine acyltransferases, which catalyze a reversible reaction involving acyl-CoA plus L-Carnitine, leading to the formation of acyl-L-Carnitine plus free CoA (Scheme 1).

It is clear from the above, that impaired activity of any of the enzymes of β-oxidation or of the auxiliary systems concerned with fatty acid transport is likely to have a major impact on lipid metabolism. The implication of L-Carnitine at this point is critical as it has been shown that disorders in L-Carnitine uptake in muscle or heart cells may lead to impaired β-oxidation resulting in myopathy or heart disease. Thus, because of its important role in fatty acid transport towards mitochondrial oxidation, L-Carnitine is a key element in fat metabolism (6).

L-CARNITINE INCREASES FATTY ACID OXIDATION

Overweight and obesity are increasing at an alarming rate in Western societies. The degree of obesity is commonly classified by the Body Mass Index (BMI = body weight / height^2). By this definition, more than 50% of U.S. adults are overweight (BMI>25), and the number of overweight children is rising steadily. L-Carnitine supplementation appears to play a role in promoting a healthy body weight when used as part of an overall weight management program which includes energy restriction and exercise. Muscle-building exercise is very important, since skeletal muscle plays a major role in whole body lipid oxidation. At rest, the oxidation of fatty acids contributes significantly to overall energy needs: up to 90% of the energy requirements of the resting muscle are obtained from fatty acid oxidation. Researchers found a marked reduction of lipid oxidation in the skeletal muscle of obese humans (7).

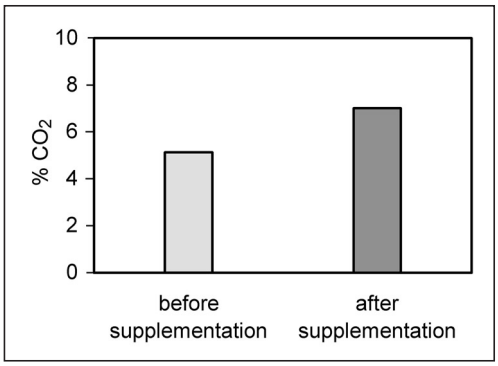


Figure 2 – Cumulative 15-hour % of $^{13}CO_2$ exhalation before and after L-Carnipure® L-Carnitine supplementation (3x1 g/day for 10 days)

The latest clinical breakthrough research comes from the Universities of Leipzig and Rostock, Germany. In 2002, researchers from the University of Leipzig published that supplementation with L-Carnipure® L-Carnitine significantly increases fatty acid oxidation (4). This study is meaningful because it is the first investigation to conclusively show that oral L-Carnipure® L-Carnitine supplementation stimulates *in vivo* long chain fatty acid metabolism in healthy

adults. The researchers investigated the effects of oral L-Carnipure® L-Carnitine supplementation on *in vivo* long chain fatty acid oxidation by measuring 1-[^{13}C] labelled palmitic acid oxidation in healthy adults before and after L-Carnipure® L-Carnitine Crystalline supplementation (3x1 g/d for 10 days). Determination of $^{13}CO_2$ was performed using an isotope-selective non-dispersive infrared spectrometry, providing test results which are comparable to measurements obtained with isotope mass spectrometry. A significant increase in $^{13}CO_2$ exhalation could be observed, indicating a significant increase in fat oxidation in healthy adults following L-Carnipure® L-Carnitine supplementation (Figure 2). According to Dr. Mueller, "this study is important to all people who exercise, those who undergo a weight management program and those who have a high energy demand".

These initial findings lately have been confirmed by another research group at the University of Rostock under the guidance of Prof. Wutzke (8). Set up to verify the first results obtained by Müller et al. by using a different approach, namely a combined ^{15}N-, ^{13}C-tracer technique, the effects of oral L-Carnipure® L-Carnitine supplementation (3x1.5 g L-Carnitine L-Tartrate/day for 10 days) on long chain fatty acid oxidation in slightly overweight adults were investigated. After oral administration of the labelled fatty acids and the amino acid glycine, ^{15}N- and ^{13}C-enrichment in breath were measured by isotope ratio mass spectrometry. The

Lonza, the leading manufacturer and supplier of the bulk dietary supplement L-Carnitine, is the only supplier who can guarantee 100% pure, natural L-Carnitine – totally free from harmful D-Carnitine. L-Carnipure® and the L-Carnipure® logo are registered trademarks of Lonza

researchers observed a significant increase in $^{13}CO_2$ exhalation after L-Carnipure® L-Carnitine supplementation, too, which again showed an increase in fatty acid oxidation.

PRODUCTION PROCESS OF L-CARNIPURE® L-CARNITINE

Carnitine, like many biologically active molecules, exists in two forms: L-Carnitine and D-Carnitine. These two forms, or isomers, are mirror images of each other. Only L-Carnitine is the naturally occurring and effective form. D-Carnitine is harmful to the human body in as it inhibits the utilisation of L-Carnitine.

For many years, the only available chemical manufacturing process had consisted of L-Carnitine separation from the racemic mixture of synthetically produced DL-Carnitine. This situation was changed when Lonza developed the synthesis of pure L-Carnitine from an optically inactive precursor molecule, using a biotransformation step for stereoselective introduction of the optical centre. Only this patented method guarantees production of pure L-Carnitine containing 0% of harmful D-Carnitine. Thus, these products are branded with the L-Carnipure® quality seal.

ADDING L-CARNIPURE® L-CARNITINE TO FUNCTIONAL FOOD

The time when L-Carnitine could only be found in red meat is well behind us.

These days, L-Carnipure® L-Carnitine can be found in sports drinks, yoghurt drinks, chewy sweets or biscuits, to mention but a few.

As the functional food and nutraceutical industry emerges from a specialised into a mainstream market, a key challenge for functional food manufacturers is to provide a product that not only delivers on performance but also on taste and texture. L-Carnipure® L-Carnitine Crystalline and L-Carnitine L-Tartrate are manufacturer-friendly. Both products are bright white, heat stable, highly water soluble and form colourless solutions.

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