Supplementation with carnitine for weight loss: a biochemical approach

José Henry Osorio, PhD*

SUMMARY

Carnitine is a molecule involved in transporting activated fatty acids among different cellular compartments, which is most likely present in all animal species, and in numerous microorganisms and plants. Recently the trend in the field of weight control is to include carnitine in the diet as an agent responsible for weight loss. In the present review, some findings are discussed from a biochemical point of view to illustrate if the use of carnitine for weight loss can be considered fiction or reality.

Keywords: Carnitine; Lipid metabolism; Weight loss.

Colomb Med. 2011; 42: 529-35

Suplementación con carnitina para perder peso: Una aproximación bioquímica

RESUMEN

La carnitina es una molécula involucrada en el transporte de ácidos grasos activados, entre diferentes compartimentos celulares, la cual posiblemente está presente en todas las especies animales, así como en numerosos microorganismos y plantas. Recientemente, la tendencia en el campo del control de peso incluye la introducción de carnitina en la dieta, como un agente responsable de la pérdida de peso. En el presente artículo de revisión, se discuten algunos hallazgos, desde un punto de vista bioquímico, para ilustrar si el uso de carnitina para la pérdida de peso puede considerarse ficción o realidad.

Palabras clave: Carnitina; Metabolismo lipídico; Pérdida de peso.

Colomb Med. 2011; 42: 529-35

Recently, an increasing number of people believe that carnitine is an important substance, which can be supplemented in diets for weight loss in humans, as well as in pets; hence, it is common to find in the marketplace a variety of products supplemented with carnitine or even pure carnitine to consume for weight loss purposes; however, many doubts remain in the field of nutrition related to the veracity of this postulate, making it necessary to delve into the metabolic ways in which this molecule is implicated.

Carnitine was discovered in muscle extracts and reported separately in 1905. Its molecular structure (3-hydroxy-4-N-trimethyl-aminobutyric acid) was established in 1927 and the name carnitine originates from the Latin word for flesh or meat, carno.

In 1952, Carter et al.1, found that carnitine was essential for the growth of the yellow mealworm, Tenebrio molitor, and it was also discovered that carnitine was present in a wide range of biological materials and that carnitine could be reversibly acetylated with acetyl-coenzyme A (CoA).

Carnitine is present in tissues and body fluids as free and as esterified short-chain, medium-chain, and long-chain acylcarnitines. Total carnitine consists of the sum of free carnitine and all acylcarnitines. Animal tissues contain relatively large amounts of carnitine, varying between 0.2 and 6 μmol/g, with the highest concentrations in heart and skeletal muscle2.

The role of carnitine in fatty acid oxidation was discovered in 1955, while working with liver homo-
genates, and the configuration of the physiological
enantiomer was determined as L(-) or R(-)-3-hydroxy-
4-N,N,N-trimethylaminobutyrate by Kaneko and
Yoshida in 1962\textsuperscript{3}. The main function of carnitine is to
shuttle activated long-chain fatty acids \([\text{fatty acyl-CoA}\] from the cytosol into the
mitochondrial matrix for \(\beta\)-oxidation, and to remove
short-chain, medium-chain, and long-chain fatty acids
that accumulate as a result of normal and abnormal
metabolism\textsuperscript{4}. Thereby, carnitine helps to maintain
adequate cellular levels of free CoA; furthermore,
products from the peroxisomal \(\beta\)-oxidation system,
including acetyl-CoA, are transported as carnitine-
esters from peroxisomes to mitochondria for complete
degradation to \(\text{CO}_2\) and \(\text{H}_2\text{O}\). Carnitine can also modulate
the toxic effects of poorly metabolised acyl-groups of
either xenobiotic origin (e.g. pivalic acid and valproate)
or those arising from various inborn errors of meta-
bolism, and it can also interact with membranes to
to change their physiochemical properties\textsuperscript{5}. This means
that carnitine modulates the acyl-CoA/free CoA ratio
via the formation of acyl-carnitines. If acyl-CoAs are
produced faster than they are utilised, intramitochondrial
free CoA is regenerated as carnitine, which binds the
acyl-groups, thus, restoring the normal intra-mito-
chondrial acyl-CoA/free CoA ratio\textsuperscript{6}.

The present review analyses the use of carnitine for
fat burning, remarking the main biochemical aspects
related to the subject. First of all, the way fat is physio-
logically burnt by the normal organism is studied, and
two more sections are included, one section shows the
sources of carnitine for humans and the destiny of
carnitine under different conditions, and the latter section
analyses the supplementation of carnitine for weight
loss.

DEGRADATION OF FATTY ACIDS

Fatty acids (FA) are stored in adipose tissue during
periods of good feeding, and they are the major source
of energy for the heart and the skeletal muscle, and one
of the most important processes for producing fuel
during endurance exercise and starvation. FA provide
as much as 80% of the energy for heart and liver
function, and the oxidation of long-chain FA also
provides the energy for non-shivering thermogenesis
by brown adipose tissue\textsuperscript{7}. The study of FA biological
degradation was done in 1904 when Knoop performed
experiments with dogs, which led him to formulatethe
theory of \(\beta\)-oxidation. This pathway is responsible for
the degradation of FA to produce acetyl-CoA, and the
mitochondrial location of this pathway agreed with the
observed coupling of FA oxidation to the citric acid
cycle and to oxidative phosphorylation\textsuperscript{8}.

Most tissues are able to degrade FA to \(\text{CO}_2\) and \(\text{H}_2\text{O}\),
but the liver has the unique capacity to synthesise
ketone bodies, acetoacetate and 3-hydroxybutyrate from
acetyl-CoA, supplying an important fuel to other organs,
mainly the brain\textsuperscript{9}.

In addition, peroxisomes and glyoxysomes,
collectively referred to as micro bodies, are sub-cellular
organelles that do not have an energy-coupled electron
transport system, but instead contain flavine oxidases,
which cata
talyse the substrate-dependent reduction of
oxygen to \(\text{H}_2\text{O}_2\). Within a few years of the identification
of peroxisomal \(\beta\)-oxidation in animals, the pathway
was elucidated and the liver enzymes had been purified
and characterised\textsuperscript{10}. FA are activated by acyl-CoA
synthetase on the peroxisomal membrane and the entry
into the organelle is independent of carnitine. Substrates
that are preferably, or exclusively, oxidised in pero-
xisomes include very long-chain FA, polyunsaturated
FA, dicarboxylic acids, prostaglandins, eicosanoids,
pristanic acid, bile acid intermediates, and side chains
of xenobiotics, which are not metabolised or poorly
metabolised by mitochondria, and the side chain of
cholesterol\textsuperscript{11}.

The mitochondrial \(\beta\)-oxidation pathway. Physiolo-
gically available FA are mostly C16 and C18 species
and include saturated and both mono- and di-unsaturated
species. The tissue uptake of FA and their transfer from
the cell membrane to the place of \(\beta\)-oxidation remain
poorly understood. FA transporters (FATP) and
cytosolic FA-binding proteins (FABP) are probably
involved in these processes\textsuperscript{12}.

For mitochondrial \(\beta\)-oxidation, long-chain FA are
activated to their CoA esters by Acyl-CoA synthase in
the cytosol and on the mitochondrial outer membrane;
the mitochondrial inner membrane is impermeable to
acyl-CoA esters. Carnitine is supplied into the cell by a
plasma membrane carnitine transporter commonly
referred to as OCTN2, located in the cellular membrane,
and three enzymes: carnitine palmitoyltransferase I
(CPTI) in the outer mitochondrial membrane; carnitine/
acylcarnitine translocase (CACT) within the inner mitochondrial membrane, and carnitine palmitoyltransferase (CPT II) in the inner mitochondrial membrane (carnitine O-palmitoyltransferase EC 2.3.1.21) are responsible for the reversible reaction: acyl-CoA+carnitine=acylcarnitine+CoA-SH. The regulation of mitochondrial FA oxidation mainly involves CPT I. In the liver, CPT I controls the FA flux through the esterification and oxidative pathways, given its sensitivity to malonyl-CoA, a potent CPT I inhibitor that is the first committed intermediate in the pathway of FA biosynthesis. During fasting, the malonyl-CoA level decreases, and CPT I becomes uninhibited, then long-chain fatty acid (LCFA) oxidation and subsequent ketogenesis become enhanced. In the post-absorptive state, the concentration of malonyl-CoA rises, CPT I is thereafter inhibited, and newly formed LCFA are directed towards esterification. Fatty Acids of less than 12 carbons, such as those provided by dietary supplements of medium-chain triglycerides, can enter the mitochondria and are activated within the mitochondrial matrix independent of the carnitine transport system.

The \(\beta\)-oxidation spiral. Once inside the mitochondria, the fatty acyl-CoA is degraded through four separate reactions. The enzyme responsible for the first step is an acyl-CoA dehydrogenase, which transfers the electrons to an electron-transfer flavoprotein (ETF) and Coenzyme Q of the respiratory chain by using a second flavoprotein ETF: CoQ oxidoreductase, also named ETF dehydrogenase.

The second step is carried out by an enoyl-CoA hydratase, and the enzyme involved in the third step is an L 3-hydroxyacyl-CoA dehydrogenase using NAD as cofactor for the reaction, which is reduced to NADH transferring electrons to complex 1 of the respiratory chain. After the fourth step in which a 3-ketoacyl-CoA thiolase is involved, cycling continues until the final thiolytic cleavage with the production of two acetyl-CoA molecules. The acetyl-CoA produced is used directly in muscle as an energy substrate through the citric acid cycle, whilst in the liver it is degraded for the production of ketone bodies as an energy source in tissues such as brain.

The distribution of the enzymes into the mitochondria is divided in membrane-bound enzymes and mitochondrial matrix enzymes.

The enzymes for the \(\beta\)-oxidation of long- to medium-chain fatty acids (C-18 to C-12) are located close to the inner mitochondrial membrane, they are: very-long-chain acyl-CoA dehydrogenase, long-chain enoyl-CoA hydratase, long-chain L 3-hydroxyacyl-CoA dehydrogenase and long-chain 3-ketoacyl-CoA thiolase. The activity for the last three enzymes is found within a multiple complex known as the mitochondrial trifunctional protein (TFP).

The enzymes located within the mitochondrial matrix are responsible for \(\beta\)-oxidation of the medium- to short-chain acyl-CoA intermediates, including long-chain, medium-chain, and short-chain acyl-CoA dehydrogenases; a short-chain hydratase, also named crotonase; a medium-/short-chain 3-hydroxyacyl-CoA-dehydrogenase; and both medium-chain and short-chain 3-keto acyl CoA thiolas.

Unsaturated FA oxidation requires auxiliary enzymes. D\(^3\),D\(^2\)-enoyl-CoA isomerase and 2,4-dienoyl-CoA reductase are the auxiliary enzymes for \(\alpha\)-oxidation of FA with double bond at an odd- and even-numbered position, respectively (linoleate isomerase E.C.5.2.1.5; cis-2-enoyl CoA reductase E.C.1.3.1.37; trans-2-enoyl CoA reductase E.C.1.3.1.38). The above-mentioned isomerase catalyses the isomerisation of both 3-cis and 3-trans-enoyl-CoAs to 2-trans-enoyl-CoAs. Enoyl-CoA isomerases for short-, medium- and long-chain substrates have been identified, and an isoform of mitochondrial 2,4-dienoyl-CoA reductase has been reported.

Sources of carnitine for humans. Carnitine synthesis in mammals is carried out from the turnover of proteins containing lysine residues, which are previously post-translationally trimethylated with the release of trimethyllysine. The rate-limiting step in the pathway is the hepatic enzyme, \(\gamma\)-butyrobetaine hydroxylase; however, the rate of carnitine biosynthesis is mainly determined by the rate of protein turnover that supplies trimethyllysin.

In humans, 98% of the carnitine resides in the skeletal and cardiac muscle with 1.6% in the liver and kidney, and 0.4% in the extra cellular fluid. Humans obtain most of their carnitine (some 50% to 75% of daily requirements) through dietary intake (meat, poultry, fish, and dairy products); with L-carnitine being primarily synthesised in the liver and also in the kidney and brain from protein-derived 6-N-trimethyllysine via 3-hydroxy-6-N-trimethyllysine, 4-N-
trimethylaminobutyraldehyde and 4-N trimethylamino-
butyrate (4-N-butyrobetaine). Other cells depend on
carnitine import via active uptake from the blood. This
transport system is also involved in the renal tubular re-
absorption and intestinal absorption of carnitine20.

There is no degradation pathway for carnitine in
mammals, although there is minor degradation of dietary
carnitine by intestinal bacteria (less than 1% to 2% in
total), and carnitine is eliminated via urine as free
carnitine and acylcarnitines with renal fractional re-
absorption of up to 90%. Carnitine uptake into tissues
and cells occurs by a saturable sodium-dependent
transport mechanism21, and a failure of its transport
mechanism leads to systemic or primary carnitine
deficiency associated with low levels of free and total
carnitine in tissues and plasma. Also, during periods of
metabolic decompensation in which acyl-CoA esters
accumulate, the concentration of acylcarnitines greatly
increases and exceeds the capacity for L-carnitine
biosynthesis (and of dietary sources), leading to a
secondary carnitine deficiency22.

Supplementation with carnitine. It has been
scientifically recognized that carnitine can be supple-
mented to improve some clinical conditions like ano-
rexia23; cardiovascular disease, angina and ischemia24;
cardiogenic shock25; cardiomyopathy26,27; myocardial
infarction28; hyperlipidemia29; insulin resistance30;
painful diabetic neuropathy31; chronic fatigue
syndrome32; fatty liver33; hepatitis and hepatic ence-
phalopathy34; immunity problems35; hyperthyroidism36;
males infertility37; renal failure/dialysis38,39; respiratory
distress in premature infants40, and inherited inborn
errors of fatty-acid oxidation41.

The levels of endogenous L-carnitine, under normal
conditions, can be influenced by long-term changes in
dietary habits and nutritional status42, some authors
pointed out that inasmuch as carnitine is not considered
a true ‘vitamin’ and, under normal conditions, healthy
humans can synthesise sufficient amounts of it43,
individuals who consumed diets low in L-carnitine
(vegetarian diets) had lower plasma L-carnitine levels
than subjects consuming a mixed diet, with children
being affected to a greater extent than adults. Despite
large variations in dietary intake, the mean plasma
levels of the compound vary by only about 20%. This
is understandable as renal excretion of L-carnitine is
substantially less in those individuals with low dietary
intake42. Therefore, carnitine biosynthesis and renal
conservation mechanisms are generally implicated and
adequate to prevent overt carnitine deficiency in
individuals who self-select diets that are low in
carnitine42.

Dietary L-carnitine intake can vary significantly
between strict vegetarians who consume less than 0.1
imol/kg/day, representing about 1 mg/day for a 70-kg
adult, and an average person with a diet providing a
daily intake of 2-12 μmol/kg/day, or 23-135 mg per day
for an average adult44; however, the extent of absorption
in the subjects fed with a low-carnitine diet may be
75%, on average45 while in subjects on a high-carnitine
diet, 37% of the dose can be accounted for as excreted
metabolites, meaning that the extent of absorption
might be about 63%. Then the efficiency of absorption
tends to diminish as the carnitine content of the diet
increases45,46, reflecting the involvement of specific
transporters that can be saturated even with normal
dietary intake. On the other hand, the bioavailability of
supplemental or medicinal oral doses of L-carnitine
tends to be even lower, at 5%-18%. Loss of endogenous
L-carnitine from the body primarily occurs via renal
excretion in the form of L-carnitine, acetyl-L-carnitine
and longer chain esters. In a 24-hour period, a healthy
human consuming a normal diet excretes between 100
and 300 μmol of total carnitine, although the overall
rate of excretion varies according to dietary intake 47.
Because L-carnitine is not bound to plasma protein48, it
is extensively filtered at the glomerulus. However,
tubular re-absorption ensures that only a small fraction
of the filtered load is excreted in urine. In healthy
individuals, the fractional tubular re-absorption of L-
carnitine (and acyl-L-carnitine derivatives) exceeds
90% and is probably greater than 98% under normal
homeostatic conditions49,50. If the tubular re-absorption
of L-carnitine is impaired due to disease or the
administration of compounds that inhibit the renal
tubular transport of the compound, the result is an
increased urinary loss, and a systemic deficiency may
develop51,52. In renal Fanconi syndrome, a significant
reduction in the tubular re-absorption of L-carnitine
results in a secondary deficiency of L-carnitine in
plasma and muscle53. In some specific cases like training
and exercise, the information obtained is somewhat
controversial, given that some authors have found
evidence for a beneficial effect of L-carnitine
supplementation during training, competition, and recovery from strenuous exercise and during regenerative athletics\textsuperscript{54}, in contrast to others\textsuperscript{55,56}, however, experimental findings support the statement that L-carnitine supplementation does not promote weight loss\textsuperscript{57-60}.

CONCLUSIONS

After detailed analysis of the biochemical means in which carnitine is implicated, it can be concluded that carnitine supplementation does not promote weight loss and that carnitine supplementation is only recommended in secondary carnitine deficiency and some inherited inborn errors based on some important positions:

- Carnitine is a very important osmolite needed to import long-chain fatty acids into mitochondria for $\beta$-oxidation. However, the homeostasis of carnitine is kept through very efficient mechanisms such as the ability of the human body to synthesize sufficient amounts of carnitine even under adverse dietetic management and a very efficient tubular re-absorption of carnitine under normal conditions.
- The carnitine cycle depends basically on a good production and an adequate function of the enzymes carnitine palmitoyltransferase I, carnitine acetyl-carnitine translocase, and carnitine palmitoyltransferase II, whose production is not stimulated by carnitine.
- Carnitine does not promote fat degradation and mobilization from adipose tissue to others tissues for energy production, as can be achieved by some hormones like glucagon.
- Negative changes in the efficiency of absorption of carnitine are present when the content of carnitine in the diet increases.

REFERENCES

22. Infante JP, Huszagh VA. Secondary carnitine deficiency and impaired docosahexaenoic (22:6n-3) acid synthesis: a common denominator in the pathophysiology of diseases of oxidative


