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Medium-Chain, Even-Numbered Dicarboxylic Acids as Novel Energy Substrates: An Update

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Medium-chain dicarboxylic acids are produced by higher plants and animals via fatty acid ω-oxidation or by β-oxidation of longer-chain dicarboxylic acids. In plants, dicarboxylic acids are components of the natural protective polymers cutin and suberin; in animals, dicarboxylic acids are mainly oxidized in mitochondria, where they are transported through four different pathways. Their energy density is intermediate between glucose and fatty acids. Dicarboxylic acid administration does not require insulin or stimulate insulin secretion, and the β-oxidation of dicarboxylic acids produces succinic acid, a gluconeogenic substrate. Therefore, dicarboxylic acids might be a suitable fuel substrate, particularly in clinical conditions in which marked insulin resistance and/or impairment of aerobic glycolysis occur.

Key words: dicarboxylic acids, fuel substrates, gluconeogenesis, nutrition

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INTRODUCTION

Sepsis, shock, multiple trauma, and burns are often associated with severe catabolism that allows wasting of lean body mass, immune dysfunction, and compromised wound healing. Nutritional support is key in the management of these critically ill patients to minimize these complications. In the early period after trauma, the metabolism slows, oxygen consumption decreases, and energy is primarily provided to the vital organs. Energy is supplied by increasing plasma glucose recruited from glycogen stores in the liver and muscle through glycogenolysis and, to a lesser extent, from protein breakdown providing amino acids for gluconeogenesis. However, within 2 to 3 days after trauma, glycogen stores are depleted and gluconeogenesis increases to fulfill energy needs using mostly amino acids from muscle. This results in depletion of muscle proteins and increased ureagenesis, leading to muscle wasting, negative nitrogen balance, loss of function of vital organs, and a delay in wound healing.[1] In the 14 days after trauma, the hypermetabolism can result in a depletion of essential protein stores, with the consequence that patients are at an increased risk of developing serious complications such as sepsis and multiple organ failure.[2]

In this situation, appropriate and optimal substrate support through parenteral and/or enteral nutrition remains of great clinical importance. Branched-chain amino acids, nonessential amino acids, acetylated amino acids, peptides, cysteine, and arginine are often used for this purpose, but prospective, randomized, controlled trials have not consistently demonstrated improved survival with their use in critically ill patients.[3] An exception seems to be glutamine, since in the catabolic state, plasma levels of glutamine are insufficient to meet increased demands.[4] Finally, glucose solutions and conventional intravenous fat emulsions have been shown to increase susceptibility to infection.[5]

Even-numbered dicarboxylic acids have been proposed as alternate energy substrates, since their β-oxidation also produces succinic acid, a gluconeogenic precursor, which could result in sparing gluconeogenic amino acid utilization. In addition, the administration of these diacids does not require insulin or stimulate insulin secretion, which is desirable because under stressful conditions, large amounts of energy might be required at a time when there are marked disturbances in glucose utilization due to insulin resistance and/or impairment of aerobic glycolysis.

This review will focus on the metabolism of medium-chain dicarboxylic acids as possible alternative energy substrates, as they represent a new class of substrates with metabolic and energetic characteristics intermediate between glucose and fatty acids.

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DICARBOXYLIC ACIDS IN PLANTS

Medium- and long-chain dicarboxylic acids are produced by both higher plants and animals via ω-oxidation of fatty acids.[6,7] In plants, dicarboxylic acids are components of the natural protective polymers cutin and suberin, which are involved in waterproofing the leaves and fruits of higher plants, regulating the flow of nutrients among various plant cells and organs, and minimizing the deleterious impact of pathogens.[8] The major components of cutin are dihydroxy C16 fatty acids, 18-hydroxy-9,10-epoxy C18 fatty acids, and trihydroxy C18 fatty acids, although smaller amounts of ω-hydroxy fatty acids from C16 to C18 are also present. The major components of suberin are ω-hydroxy fatty acids and dicarboxylic acids with a chain length from 16 to 28 carbon atoms and alcohols with a chain length generally longer than C20. There is a large body of literature showing that when aerial portions of the plants such as leaves or fruits are wounded, they produce cutin or suberin to seal off the wound.[8]

Dicarboxylic acids are β-oxidized in plant peroxisomes. Glyoxysomes are specialized peroxisomes present in various plant organs such as germinating cotyledons and senescing leaves. They are the site of β-oxidation and of the glyoxylate cycle, pathways essential to maintaining the gluconeogenesis deriving from degradation of reserve or structural lipids.[9]

DICARBOXYLIC ACIDS IN ANIMALS AND HUMANS

Omega Oxidation

In animals and humans, medium-chain dicarboxylic acids, which include adipic (C6), suberic (C8), sebacic (C10), and dodecanedioic (C12) acids (Figure 1), derive from the β-oxidation of longer-chain dicarboxylic acids.[10] Long-chain dicarboxylic acids, in turn, are formed from the correspondent fatty acids by ω-oxidation (Figure 2) in the microsomial membranes[11] or are consumed with a diet rich in vegetables. However, there is evidence of a direct conversion of lauric acid to dodecanedioic acid—that is, from a monocarboxylic acid with 12 carbon atoms to the correspondent C12 dicarboxylic acid. In fact, when the recombinant fusion protein rF450[mRat4Al/mRatOR]L1, containing the heme domain of P450 4A1 and the flavin domains of NADPH-P450 reductase, is incubated with dilaurylphosphatidylcholine, it catalyzes the ω-oxidation of lauric acid at a very high rate (about 300 nmol/min/nmol P450).[12] The ω-oxidative pathway is prominent in brain and could play a role in brain fatty acid metabolism. In fact, Alexander et al.[13] showed that homogenates of rat brain catalyze the ω-oxidation of monocarboxylic acids with a specific activity of between 0.87 and 5.23 nmol/mg of post-mitochondrial protein per hour, depending on the substrate. This activity is remarkably higher (0.25 to 4 times) than that found in rat liver, depending on the chain length of the substrate. Specific activity increases with increasing chain length of the substrate. Cultured rat neurons, astrocytes, and oligodendrocytes all contain ω-oxidation activity, and the product of ω-oxidation in the brain is almost exclusively dicarboxylic acid.[13]

ADIPIC ACID	HOOC—$(CH_2)_4$—COOH
SUBERIC ACID	HOOC—$(CH_2)_6$—COOH
SEBACIC ACID	HOOC—$(CH_2)_8$—COOH
DODECANEDIOIC ACID	HOOC—$(CH_2)_{10}$—COOH

Figure 1. Chemical structure and nomenclature of the main medium-chain dicarboxylic acids.

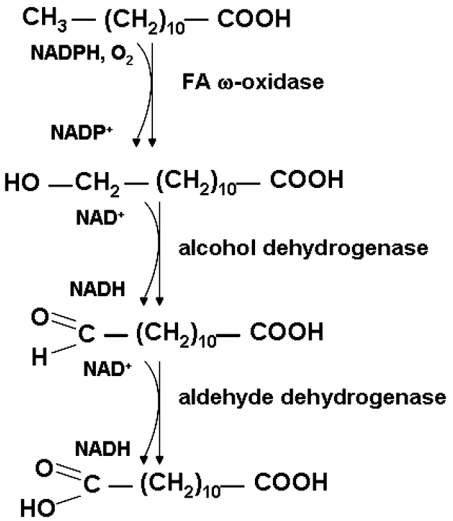

Figure 2. Fatty acid ω-oxidation. The ω-methyl group of a fatty acid (e.g., dodecanoic or lauric acid) is first oxidized to an alcohol group, and is then further oxidized to form an aldehydic group and finally a carboxylic group. At the end of these oxidative steps, a fatty acid is converted into the correspondent dicarboxylic acid.

Gluconeogenic Properties of Dicarboxylic Acids

It has been proposed by Mortensen[14] that fatty acid ω-oxidation might play a physiologic anti-ketogenic and gluconeogenic role in energy metabolism. In fact, we have shown that after administering adipic acid to ketotic

rats, urinary excretion of succinic acid increased at the same time as ketosis decreased and blood glucose increased.

As glucose is an essential nutrient for the human body, its circulating levels must be carefully maintained to supply adequate amounts to peripheral tissues. The liver plays a central role in glucose homeostasis by balancing its uptake and storage via glycogenesis and its release via glycogenolysis and gluconeogenesis. An impairment of glucose homeostasis is a typical feature of type 2 diabetes. In fact, type 2 diabetic patients exhibit a marked reduction of the net hepatic glycogen synthesis after either a mixed meal or during combined hyperinsulinemic hyperglycemia.[15]

Glycogen synthesis is a major pathway of glucose disposal in skeletal muscle and is regulated by the insulin-sensitive and rate-limiting enzyme glycogen synthase. Defects in this enzyme can significantly alter the intracellular routing and metabolism of glucose and contribute to insulin resistance in muscle tissue. Skeletal muscle glycogen synthase has been shown to be scarcely stimulated by insulin in Caucasian subjects with type 2 diabetes and in Pima Indians,[16,17] as well as in relatives of patients with diabetes,[18] demonstrating their insulin resistance.

In other pathological conditions associated with hypercatabolism, such as sepsis and burns, sources of energy are provided by gluconeogenesis, lipolysis, and ketogenesis. However, sepsis is associated with abnormality of virtually all of these provisional sources of energy substrates[18] because it impairs the function of the glycolytic pathway, the integrity of which is necessary for glucose to be used effectively to produce energy.[18]

When glucose homeostasis is impaired, as demonstrated by a reduction of its peripheral utilization, alternate fuel substrates become essential. In fact, while de novo lipogenesis, the enzymatic pathway for converting dietary carbohydrate into fat, is available in humans, the capacity to convert fats into carbohydrates does not exist. Therefore, during higher catabolism, amino acids are preferentially utilized by the liver. In these situations, amino acids are the major fuel of liver—in fact, their oxidative conversion to glucose accounts for about one-half of the daily oxygen consumption of the liver, more than any other fuel.[19] The daily supply of amino acids provided in the diet cannot be totally oxidized to carbon dioxide in the liver, because such a process would provide far more ATP than the liver could utilize. Instead, most amino acids are oxidatively converted to glucose. This results in an overall ATP production during amino acid oxidation very nearly equal to the ATP required to convert amino acid carbon to glucose.

The energy cost of gluconeogenesis is as follows:

$$2 \text{ pyruvate} + 4\text{ATP} + 2\text{GTP} + 2\text{NADH} + 2\text{H}^+ + 4\text{H}_2\text{O} \rightarrow \text{glucose} + 2\text{NAD}^+ + 4\text{ADP} + 2\text{GDP} + 6\text{Pi}$$

where for each molecule of glucose there are six high-energy phosphorus molecules used and two molecules of NADH. Overall, the "cost" of making glucose from pyruvate is two ATP equivalents (i.e. –2 ATP/mole pyruvate). In contrast, the synthesis of glucose from succinic acid (Tables 1 and 2) deriving from the oxidation of even-numbered dicarboxylic acids, is +1 ATP, or 7.3 kcal/mole succinate.

The use of even-numbered dicarboxylic acids can supply succinic acids to both liver and muscle, thus increasing glunoneogenesis (Figure 3), particularly in those clinical conditions in which glycogen stores are inadequate, for example, in diabetes and in sepsis.

Beta-Oxidation of Dicarboxylic Acids

The oxidation of dicarboxylates can take place in both mitochondria and peroxisomes.[20,21] However, until 1996 the mechanism of transport of these acids into peroxisomes and mitochondria was not established.[20,21] Liu et al.[25] demonstrated that four different pathways for the transport of dicarboxylates operate in mitochondria. These pathways include electrophoretic transport via the inner membrane anion channel, electroneutral transport, tributyltin-mediated transport, and transport via the dicarboxylate carrier. The latter can transport only oxalate, malonate, and succinate, while dicarboxylic acids with longer chain length enter the mitochondria via the other transport mechanisms.

Palmitate and medium-chain dicarboxylic acids (C10 and C12) enter the mitochondria through different processes. Unlike palmitate, C10 and C12 do not depend upon transport across the cell membrane by the fatty acid transporters, activation in the extra-mitochondrial space, or upon transport into the matrix by the carnitine shuttle (CPT1, CPT2, and CAT). Consequently, while LCFA utilization may be tightly regulated by the activity of the carnitine shuttle, C10 and C12 bypass this control network (Figure 4).

Dicarboxylic Acids in Nutrition

Medium-chain dicarboxylic acids, those with a chain length from 6 to 12 carbon atoms, have been recently regarded as possible alternate fuel substrates in both normal and pathological conditions in man, since they are rapidly β-oxidized in both mitochondria and peroxisomes.[25,27] Even-chain dicarboxylic acids such as sebacic acid (C10) and dodecanedioic acid (C12) pro-

Table 1. Energy Metabolism of Sebacic Acid

Reaction	ATP*
β-Oxidation	
Sebacic acid + 3FAD + 3NAD^+ + 3CoA + H_2O ⇛ Succinyl-CoA + 3Acetyl-CoA + 3$FADH_2$ + 3NADH + 3H^+	13†
3Acetyl-CoA + 3FAD + 9NAD^+ + 3GDP + 3P_i + 6H_2O ⇛ 6CO_2 + 3$FADH_2$ + 9NADH + 3GTP° + 6H^+ + 3CoA	36
Succinyl-CoA ⇛ ⇛ ⇛ 2 CO_2 + $FADH_2$ + 3NADH + GTP° + 2H^+ + CoA	12
NET:	61
Gluconeogenesis	
Succinyl-CoA + Pi + GDP ⇔ Succinate + GTP + CoA-SH	1
Succinate + E-FAD ⇔ Fumarate + E-$FADH_2$	2
Fumarate + H_2O ⇔ L-Malate	0
L-Malate + NAD^+ ⇔ Oxaloacetate + NADH + H^+	3
Oxaloacetate + GTP ⇔ phosphoenolpyruvate + CO_2 + GDP	−1
phosphoenolpyruvate + H_2O ⇔ 2-phosphoglycerate	0
2-phosphoglycerate ⇔ 3-phosphoglycerate	0
3-phosphoglycerate + ATP ⇔ 1,3-diphosphoglycerate + ADP	−1
1,3-diphosphoglycerate + NADH + H^+ ⇔ 3-phosphoglyceraldehyde + NAD^+ + P_i	−3
3 phosphoglyceraldehyde ⇔ dihydroxyacetonephosphate	0
3-phosphoglyceraldehyde + dihydroxyacetonephosphate ⇔ fructose-1,6-diphosphate	0
fructose-1,6-diphosphate + H_2O ⇔ fructose-6-phosphate + P_i	0
fructose-6-phosphate ⇔ glucose-6-phosphate + P_i	0
glucose-6-phosphate + H_2O ⇔ glucose + P_i	0
NET:	1

*One turn of the citric acid cycle generates 12 ATP:

- One high-energy phosphate (° GTP + ADP ⇔ GDP + ATP)
- Three NADH ⇛ 1 NADH produces 3 ATP
- One $FADH_2$ ⇛ 1 $FADH_2$ produces 2 ATP

†Two ATP are used in the activation of dicarboxylic acid.

duce as an intermediate metabolite succinyl-CoA, a gluconeogenic substrate that might play a relevant role in those clinical conditions in which glucose metabolism is impaired, such as starvation, sepsis, and diabetes.[28]

The transmembrane transport of dicarboxylic acid seems to be mediated by a carrier, which has been characterized in rats in the cellular membranes of both hepatocytes[29] and renal tubular cells[30,31] rather than in mitochondria.[32]

Among medium-chain dicarboxylic acids, C12 seems to be the most suitable for nutritional purposes. In fact, the urinary excretion of C12 is low (3%–5% of administered dose)[33,34] compared with azelaic acid (dicarboxylic acid with nine carbon atoms)[35] and sebacic acid (dicarboxylic acid with 10 carbon atoms),[36,37] and the energy density is high (7.18 kcal/g of C12 oxidized).[33] The C12 respiratory quotient (0.77) is rather low, representing an advantage in patients with respiratory distress, in whom CO_2 pulmonary exchange is low with subsequent hypercapnia and acidosis. Furthermore, the free fraction of C12 in plasma is higher than the fraction of both long- and medium-chain monocarboxylic acids, in view of its relatively high water solubility and its low affinity for albumin-binding sites.[38] Finally, unlike both long- and medium-chain monocarboxylic acids, C12 administered in free form as a salt does not require hydrolysis before cellular utilization.

The disposition of C12 in humans has been investigated[39] by a mathematical model also taking into account the kinetics of CO_2 produced by C12 oxidation. In this study the maximal rate of C12 tissue uptake estimated by the model was 0.38 ± 0.08 mmol/min, with a maximal calorie delivery of 750 kcal/d.

By using the euglycemic hyperinsulinemic clamp technique to evaluate insulin sensitivity, it has been demonstrated[40] that sebacate (C10) administration in healthy volunteers was associated with a glucose-sparing effect.

Furthermore, C12 infusion in type 2 diabetic patients decreased plasma glucose levels to a normal range without influencing plasma insulin levels.[41] These findings suggest that in type 2 diabetes, in which both glucose oxidation and storage as glycogen are impaired,[42,43] C12 as an alternate substrate, thanks to its intermediate biochemical and metabolic characteristics between free fatty acids and glucose, might be useful

Table 2. Energy Metabolism of Dodecanedioic Acid

Reaction	ATP*
β-Oxidation	
Dodecanedioic acid + 4FAD + $4NAD^+$ + 4CoA + H_2O ⇛ Succinyl-CoA + 4Acetyl-CoA + $4FADH_2$ + 4NADH + $4H^+$	18†
4Acetyl-CoA + 4FAD + $12NAD^+$ + 4GDP + $4P_i$ + $8H_2O$ ⇛ $8CO_2$ + $4FADH_2$ + 12NADH + 4GTP° + $8H^+$ + 4CoA	48
Succinyl-CoA ⇛ ⇛ ⇛ 2 CO_2 + $FADH_2$ + 3NADH + GTP° + $2H^+$ + CoA	12
NET:	78
Gluconeogenesis	
Succinyl-CoA + Pi + GDP ⇔ Succinate + GTP + CoA-SH	1
Succinate + E-FAD ⇔ Fumarate + E-$FADH_2$	2
Fumarate + H_2O ⇔ L-Malate	0
L-Malate + NAD^+ ⇔ Oxaloacetate + NADH + H^+	3
Oxaloacetate + GTP ⇔ phosphoenolpyruvate + CO_2 + GDP	−1
phosphoenolpyruvate + H_2O ⇔ 2-phosphoglycerate	0
2-phosphoglycerate ⇔ 3-phosphoglycerate	0
3-phosphoglycerate + ATP ⇔ 1,3-diphosphoglycerate + ADP	−1
1,3-diphosphoglycerate + NADH + H^+ ⇔ 3-phosphoglyceraldehyde + NAD^+ + P_i	−3
3 phosphoglyceraldehyde ⇔ dihydroxyacetonephosphate	0
3-phosphoglyceraldehyde + dihydroxyacetonephosphate ⇔ fructose-1,6-diphosphate	0
fructose-1,6-diphosphate + H_2O ⇔ fructose-6-phosphate + P_i	0
fructose-6-phosphate ⇔ glucose-6-phosphate + P_i	0
glucose-6-phosphate + H_2O ⇔ glucose + P_i	0
NET:	1

*One turn of the citric acid cycle generates 12 ATP:

- One high-energy phosphate (° GTP + ADP ⇔ GDP + ATP)
- Three NADH ⇛ 1 NADH produces 3 ATP
- One $FADH_2$ ⇛ 1 $FADH_2$ produces 2 ATP

†Two ATP are used in the activation of dicarboxylic acid.

because it is promptly oxidized in high amounts and supplies succinic acid for gluconeogenesis.

Recent studies have described the oxidation and the insulinotropic action of C12 in isolated pancreatic islets prepared from normal rats.[44,45] The energy density of medium-chain dicarboxylic acids (4.97 kcal/g for azelaic acid, C9; 6.64 kcal/g for sebacic acid, C10; and 7.2 kcal/g for dodecanedioic acid, C12) is roughly in between the values known for the conventional energy substrates such as glucose (3.75 kcal/g) and fatty acids (9 kcal/g).

Indeed, the physical-chemical characteristics of medium-chain, even-numbered dicarboxylic acids—their solubility in water and their gluconeogenic properties—put these molecules in a separate class between fat and carbohydrates.

Energy Profile of Dicarboxylic Acids

The equation for the complete degradation of palmitate (C16:0) is:

palmitoyl CoA + 7FAD + 7NAD + 7CoA + $7H_2O$ → 8acetyl CoA + 7FADH2 + 7NADH + $7H^+$

Palmitate oxidation yields the following:

8 acetyl CoAs enter the citric acid cycle and give:

- 24 NADH = 72 ATP (by oxidative phosphorylation)
- 8 FADH2 = 16 ATP (by oxidative phosphorylation)
- 8 GTP = 8 ATP
- 7 NADH generated by β-oxidation itself = 21 ATP (by oxidative phosphorylation)
- 7 FADH2 generated by β-oxidation itself = 14 ATP (by oxidative phosphorylation)

Total number of ATP from 1 molecule of palmitate = 72 + 16 + 8 + 21 + 14 = 131.

High-energy phosphate bonds, the equivalent of 2 ATP, are used to activate palmitate to palmitoyl CoA. Therefore, the ATP yield is 129, which in terms of energy is 941.7 kcal/mole of palmitate. The oxidation of sebacic acid and that of dodecanedioic acid are summarized in Tables 1 and 2, respectively. The ATP yield of 1 mole sebacic acid oxidation is 61 ATP, corresponding to 445.3 kcal/mole. The ATP yield of 1 mole dodecanedioic acid oxidation is 78 ATP, corresponding to 569.4 kcal/mole.

Therefore, the energy liberated during the oxidation

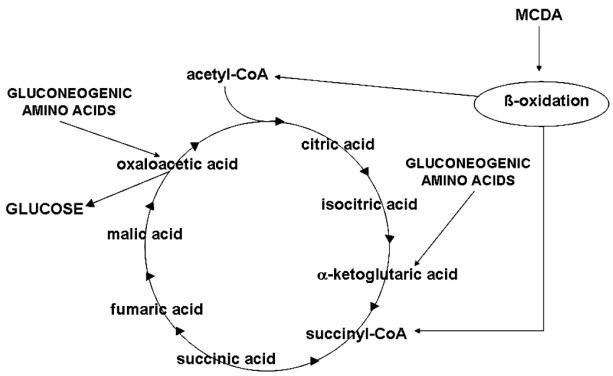

Figure 3. β-oxidation of medium-chain dicarboxylic acids and their effect on amino acid sparing in the gluconeogenic pathway. Aspartate and asparagine oxidation produce oxaloacetate; arginine, glutamate, glutamine, histidine, and proline produce alpha-ketoglutarate; isoleucine, methionine, and valine produce succinyl-CoA; and phenylalanine and trosine produce fumarate. All of these substrates are precursors of pyruvate and thus contribute to gluconeogenesis. Even-numbered, medium-chain dicarboxylic acids, through their end product, succinyl-CoA, produce oxaloacetate and thus ultimately glucose.

of 1 mole of C10 is about 47% and the oxidation of 1 mole of C12 is about 60% of the energy delivered by the oxidation of 1 mole of palmitate.

Overall, the net reaction of glucose complete oxidation is:

$$6O_2 + C_6H_{12}O_6 + 38ADP \rightarrow 38ATP + 6CO_2 + 6H_2O$$

In terms of energy yield, the oxidation of 1 mole of glucose produces 277.4 kcal, corresponding to approximately 62% and 49% of the energy produced by 1 mole C10 oxidation and 1 mole C12 oxidation, respectively.

DICARBOXYLIC ACID TRIGLYCERIDES

To reduce the amount of inorganic salts (sodium, potassium, calcium, and magnesium) administered together with dicarboxylic acids to render their molecules hydrosoluble, esters of dicarboxylic acids with glycerol, as triglycerides, have been synthesized. In this way, the amount of inorganic cations administered is halved.

The triglyceride of dodecanedioic acid, while preserving its water solubility, appears to have the lowest urinary excretion in experimental animals, corresponding to approximately 0.67% of the administered amount. Kinetic studies in rats[46] have shown that this dicarboxylic acid-C12 triglyceride has a large volume of distribution (approximately 0.5 L/kg body wt) and a fast disappearance rate from plasma (0.42/min), whereas its product of hydrolysis, the dicarboxylic acid C12, has a very small volume of distribution (approximately 0.04 L/kg body wt) and a high tissue uptake, with a maximal transport rate of 0.636 mM/min. The measured dicarboxylic acid-C12 tissue uptake rate is consistent with the possibility of achieving substantial energy delivery when added to parenteral nutrition formulations. Finally, the amount of sodium administered with the triglyceride form is one-half of that necessary for water solubility with the free diacid.

DICARBOXYLIC ACIDS REDUCE FATIGUE DURING PHYSICAL EXERCISE

Metabolically healthy skeletal muscle has the ability to switch easily between glucose and fat oxidation in response to homeostatic signals. In type 2 diabetes mellitus and obesity, the skeletal muscle shows a great reduction in this metabolic flexibility.[47] In type 2 diabetic patients, irrespective of their treatment status, there is an increased tendency for muscle to become fatigued.[48] It is believed that inefficient ATP production in response to continued high demand is a factor in muscle performance. Therefore, a decline in muscle performance, often described as muscle fatigue, is observed

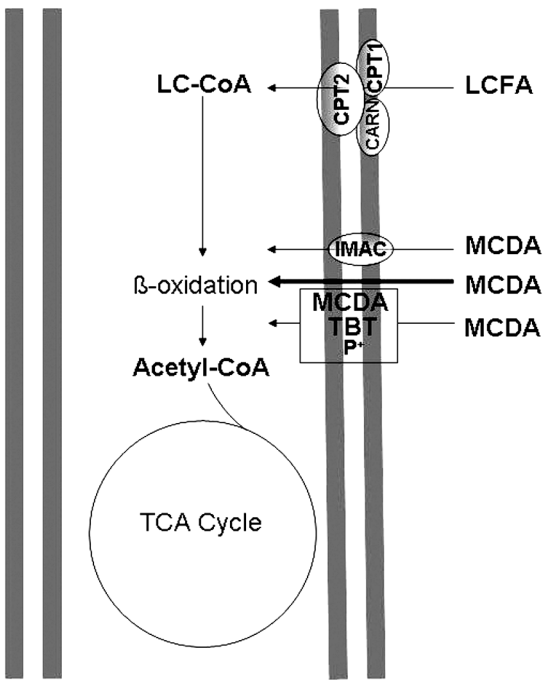

Figure 4. Schematic representation of the mechanisms of transportation of long-chain fatty acids (LCFA) and medium-chain dicarboxylic acids (MCDA) into the mitochondrion. LCFAs require the carnitine (CARN) transport system, which couples the two enzymes CPT1 and CPT2, respectively located on the outer and the inner mitochondrial membrane. MCDAs are transported via a mitochondrial inner membrane anion channel (IMAC), via passive diffusion (large arrow), which is gradient related, and through a third transport mechanism, which requires the presence of tributyltin (TBT). TBT complexes one end of the dicarboxylate and a proton (P+) binds to the other, thus forming a lipophilic neutral complex that can cross the bilayer.

when ATP production cannot be sustained during continuous activity. Therefore, muscle fatigue might be a consequence of an impaired ATP synthesis in response to a higher request, possibly secondary to a reduced mitochondrial function, largely described in type 2 diabetes.[49] Magnetic resonance spectroscopy studies in humans suggest that a defect in insulin-stimulated glucose transport in skeletal muscle is the primary metabolic abnormality in insulin-resistant patients with type 2 diabetes.[50] Excess fatty acids appear to cause this defect of glucose transport by inhibiting insulin-stimulated tyrosine phosphorylation of insulin receptor substrate-1 and its associated phosphatidylinositol 3-kinase activity.[50]

The ingestion of 40 g of C12 in type 2 diabetic subjects before moderate exercise reduces muscle fatigue, thus allowing completion of the exercise, and does not promote insulin secretion; rather, it is associated with an increase in triglyceride hydrolysis, as shown by the significant rise of plasma non-esterified fatty acids (NEFA). Forty-seven percent of C12 is taken up by peripheral tissues, and 69% of this amount is oxidized in the observation period, while the remaining fraction (53%) is possibly taken up by the liver to synthesize glycogen. In contrast, the ingestion of 40 g of C12 in healthy subjects before moderate exercise allows a C12 uptake of 33.4 g, corresponding to 83% of its intake, of which 26.7 g (80% of the amount taken up by tissues) is oxidized in the observation period.[51]

In type 2 diabetes, the incapacity to shift from glucose to lipids and vice versa as an energy substrate, depending on the energy requirement, is defined as "metabolic inflexibility." C12 may overcome this impairment by providing intermediate substrates for mitochondrial oxidation and ATP synthesis.

CONCLUSIONS

Dicarboxylic acids, and in particular dodecanedioic acid and the sodium salt of its triglyceride, seem to be a suitable alternative energy substrate for a number of reasons, including their low cost, peripheral venous administration, high energy delivery, and gluconeogenic and glucose-sparing effects.

REFERENCES

1. Wilmore DW. Alterations in protein, carbohydrate and fat metabolism in injured and septic patients. J Am Coll Nutr. 1983;2:3–13.
2. Wapnir RA, Lifshitz F. Fasting-induced hypoglycemia in experimentally malnourished rats. J Nutr. 1977;107:383–390
3. Atkinson M, Worthley LI. Nutrition in the critically ill patient: part II. Parenteral nutrition. Crit Care Resusc. 2003;5:121–136
4. Boelens PG, Nijveldt RJ, Houdijk APJ, et al. Glutamine alimentation in catabolic state. J Nutr. 2001; 131:2569S–2577S
5. Hawker FH. How to feed patients with sepsis. Curr Opin Crit Care. 2000;6:247–252.
6. Verkade PE, Van der Lee J. Researches on fat metabolism II. Biochem J. 1934;28:31–40.
7. Kolattukudy PE, Walton TJ, Kushwaha RP. Biosynthesis of the C18 family of cutin acids: omega-hydroxyoleic acid, omega-hydroxy-9,10-epoxystearic acid, 9,10,18-trihydroxystearic acid, and their delta12-unsaturated analogs. Biochemistry. 1973; 12:4488–4498.
8. Kolattukudy PE. Polyesters in higher plants. Adv Biochem Eng Biotechnol. 2001;71:1–49.
9. Penfield S, Graham S, Graham IA. Storage reserve mobilization in germinating oilseeds: Arabidopsis as a model system. Biochem Soc Trans. 2005;33:380–383.
10. Kolvraa S, Gregersen N. In vitro studies on the oxidation of medium-chain dicarboxylic acids in rat liver. Biochim Biophys Acta. 1986;876:515–525.
11. Nilsson A, Arey H, Pedersen JI, et al. The effect of high-fat diets on microsomal lauric acid hydroxylation in rat liver. Biochim Biophys Acta. 1986;879: 209–214.
12. Shet M, Fisher CW, Holmans PL, et al. The omega-hydroxlyation of lauric acid: oxidation of 12-hydroxlauric acid to dodecanedioic acid by a purified recombinant fusion protein containing P450 4A1 and NADPH-P450 reductase. Arch Biochem Biophys. 1996;330:199–208.
13. Alexander JJ, Snyder A, Tonsgard JH. Omega-oxidation of monocarboxylic acids in rat brain. Neurochem Res. 1998;23:227–233.
14. Mortensen PB. The possible antiketogenic and gluconeogenic effect of the omega-oxidation of fatty acids in rats. Biochim Biophys Acta. 1980;620:177–185.
15. Krssak M., Brehm A., Bernroider E. Alterations in postprandial hepatic glycogen metabolism in type 2 diabetes. Diabetes. 2004;53:3048–3056.
16. Bak JK, Moller N, Schmitz O, et al. In vivo insulin action and muscle glycogen synthase activity in type II (non-insulin-dependent) diabetes mellitus: effects of diet treatment. Diabetologia. 1992;35:777–784.
17. Bogardus C, Lillioja S, Mott D, et al. Relationship between obesity and maximal insulin-stimulated glucose uptake in vivo and in vitro in Pima indians. J Clin Invest. 1984;73:800–805
18. Vaag A, Henriksen JE, Beck-Nielsen H. Decreased insulin activation of glycogen synthase in skeletal muscles in young nonobese Caucasian first degree relatives of patients with non-insulin dependent diabetes mellitus. J Clin Invest. 1992;89:782–788
19. Crouser ED, Dorinsky PM. Metabolic consequences of sepsis. Correlation with altered intracellular calcium homeostasis. Clin Chest Med. 1996;17:249–261.
20. Jungas RL, Halperin ML, Brosnan JT. Quantitative analysis of amino acid oxidation and related gluconeogenesis in humans Physiol Rev. 1992;72:419–448.
21. Mortensen, P.B. C6–C10-dicarboxylic aciduria in starved, fat-fed and diabetic rats receiving decanoic acid or medium-chain triacylglycerol. An in vivo

measure of the rate of beta-oxidation of fatty acids. Biochim Biophys Acta. 1981;664:349–355.
22. Mortensen PB, Gregersen N. The biological origin of ketotic dicarboxylic aciduria. In vivo and in vitro investigations of the omega-oxidation of C6-C16-monocarboxylic acids in unstarved, starved and diabetic rats. Biochim Biophys Acta. 1981;666:394–404.
23. Passi S, Picardo M, Nazzaro-Porro M, et al. Azelaic acid–biochemistry and metabolism. Biochem Pharmacol. 1984;33:103–108.
24. Robins EJ, Breathnach AS, Bennett D, Ward BJ, et al. Ultrastructural observations on the effect of azelaic acid on normal human melanocytes and a human melanoma cell line in tissue culture. Br J Dermatol. 1985;113:687–697.
25. Liu G, Hinch B, Beavis AD. Mechanisms for the transport of alpha,omega-dicarboxylates through the mitochondrial inner membrane. J Biol Chem. 1996;271:25:338–344.
26. Sholtz KF, Bondarenko DI, Mamaev DV. Probing the active center of the mitochondrial dicarboxylate transporter. FEBS Lett. 1993;327:54–56.
27. Mortersen PB, Kølvraa S, Gregersen N. Cyanide insensitive and clofibrate enhanced β-oxidation of dodecanedioic acid: evidence of peroxisomal β-oxidation of dicarboxylic acids. Biochim Biophys Acta. 1982;713:393–397.
28. Greco AV, Mingrone G. Dicarboxylic acids, an alternate fuel substrate in parenteral nutrition: an update. Clin Nutr. 1995;14:143–148.
29. Boelsterli UA, Zimmerli B, Meier PJ. Identification and characterization of a basolateral dicarboxylate/cholate antiport system in rat hepatocytes. Am J Physiol. 1995;268:G797–G805.
30. Sheridan E, Rumrich G, Ullrich KJ. Reabsorption of dicarboxylic acids from the proximal convolution of rat kidney. Pfluegers Arch. 1983;399:18–28.
31. Ullrich KJ, Fasold H, Rumrich G, et al. Secretion and contraluminal uptake of dicarboxylic acids in the proximal convolution of rat kidney. Pfluegers Arch. 1984;400:241–249.
32. Saint-Macary M, Foucher B. Comparative partial purification of the active dicarboxylate transport system of rat liver, kidney and heart mitochondria. Biochem Biophys Res Commun. 1985;133:498–504.
33. Mingrone G, Greco AV, De Gaetano A, et al. Pharmacokinetic profile of dodecanedioic acid, a proposed alternative lipid substrate for parenteral nutrition. J Parenter Enteral Nutr. 1994;18:225–230.
34. Bertuzzi A, Mingrone G, Gandolfi A, et al. Pharmacokinetic analysis of dodecanedioic acid in humans from bolus data. J Parenter Enteral Nutr. 1995;19:498–501.
35. Bertuzzi A, Gandolfi A, Salinari S, et al. Pharmacokinetic analysis of azelaic acid disodium salt. Clin Pharmacokinet. 1991;20:411–419.
36. Mingrone G, Greco AV, Bertuzzi A, et al. Tissue uptake and oxidation of disodium sebacate in man. J Parenter Enteral Nutr. 1991;15:454–459.
37. Bertuzzi A, Gandolfi A, Salinari S, et al. Pharmacokinetics of dicarboxylic acids in man. IEEE Eng Med Biol. 1994;13:472–478.
38. Bertuzzi A, Mingrone G, Gandolfi A, et al. Pharmacokinetic analysis of dodecanedioic acid in humans from bolus data. J Parenter Enteral Nutr. 1995;19:498–501.
39. Bertuzzi A, Mingrone G, Gandolfi A, et al. Disposition of dodecanedioic acid in humans. J Pharmacol Exp Ther. 2000;292:846–52.
40. Raguso CA, Mingrone G, Greco AV, et al. Dicarboxylic acids and glucose utilization in humans: effect of sebacate. JPEN J Parenter Enteral Nutr. 1994;18:9–13.
41. Greco AV, Mingrone G, Capristo E, et al. The metabolic effect of dodecanedioic acid infusion in non-insulin-dependent diabetic patients. Nutrition. 1998;14:351–357.
42. Falholt K, Jensens I, Lindkaer Jensen S, et al. Carbohydrate and lipid metabolism of skeletal muscle in type 2 diabetes patients. Diabet Med. 1988;5:27–31.
43. Beck-Nielsen H. Insulin resistance in skeletal muscles of patients in diabetes mellitus. Diabetes Metab Rev. 1989;5:487–493.
44. Malaisse WJ, Greco AV, Mingrone G. Effects of aliphatic dioic acids and glycerol-1,2,3-tris(dodecanedioate) on D-glucose-stimulated insulin released in rat pancreatic islets. Br J Nutr. 2000;84:733–736.
45. Malaisse WJ, Greco AV, Mingrone G. Oxidation of [1,12-14C]dodecanedioic acid by rat pancreatic islets. Int J Mol Med. 2000;6:453–454.
46. Panunzi S, De Gaetano A, Mingrone G. Approximate linear confidence and curvature of a kinetic model of dodecanedioic acid in humans. Am J Physiol Endocrinol Metab. 2005;289:E915–E922.
47. Kelley DE. Skeletal muscle fat oxidation: timing and flexibility are everything J Clin Invest. 2005;115:1699–1702.
48. Halvatsiotis P, Short KR, Bigelow M, Nair KS. Synthesis rate of muscle proteins, muscle functions, and amino acid kinetics in type 2 diabetes. Diabetes. 2002;51:2395–2404.
49. Stumvoll M, Goldenstein BJ, van Haeften TW. Type 2 diabetes: principles of pathogenesis and therapy. Lancet. 2005;365:1333–1346.
50. Parish R, Petersen KF. Mitochondrial dysfunction and type 2 diabetes. Curr Diab Rep. 2005;5:177–183.
51. Salinari S, Bertuzzi A, Gandolfi A, et al. Dodecanedioic acid overcomes metabolic inflexibility. Am J Physiol Endocrinol Metab. 2006; In press.