FOREWORD

The Life Sciences Research Office (LSRO), Federation of American Societies for Experimental Biology (FASEB), provides scientific assessments of topics in the biomedical sciences. Reports are based upon comprehensive literature reviews and the scientific opinions of knowledgeable investigators engaged in work in specific areas of biology and medicine.

This technical report was developed for the Food and Drug Administration (FDA) in accordance with the provisions of Contract No. FDA 223-79-2275. It was prepared and edited by Peggy R. Borum, Ph.D., Associate Professor, Department of Food Science and Nutrition, University of Florida, Gainesville, Florida and Kenneth D. Fisher, Ph.D., Director, LSRO, FASEB.

The LSRO acknowledges the contributions of the investigators and consultants who assisted with this study. The report reflects the opinions expressed by an ad hoc study group that met at the Federation on June 14-15, 1982. The study participants reviewed a draft of the report and their various viewpoints were incorporated into the final report. The study participants and LSRO accept responsibility for the accuracy of the report; however, the listing of these individuals in Section X does not imply that they specifically endorse each study conclusion.

The report was reviewed and approved by the LSRO Advisory Committee (which consists of representatives of each constituent Society of FASEB) under authority delegated by the Executive Committee of the Federation Board. Upon completion of these review procedures, the report was approved and transmitted to FDA by the Executive Director, FASEB.

While this is a report of the Federation of American Societies for Experimental Biology, it does not necessarily reflect the opinion of each individual member of the FASEB constituent Societies.

November 27, 1983

Kenneth D. Fisher, Ph.D.
Director
Life Sciences Research Office
SUMMARY

This report summarizes the discussions of an ad hoc group that reviewed the health effects of dietary carnitine. It focuses on the current state of knowledge regarding dietary sources, essentiality, differential effects of the optical isomers, and consequences of oral administration of carnitine.

Metabolic requirements for carnitine are met both by consumption of foods containing carnitine and endogenous synthesis in the liver and kidney. Foods of animal origin are the primary source of dietary carnitine. Dietary L-carnitine and DL-carnitine appear to be absorbed in the small intestine by an active transport system and by passive transluminal absorption. Dietary L-carnitine is transported to liver and kidney as well as cardiac and skeletal muscle in healthy adults. Exogenous and endogenous carnitine appears to be transported by specific carrier systems. Cellular influx and efflux of carnitine are carrier-mediated processes in which the D-isomer is a competitive inhibitor of the L-isomer.

While L-carnitine is essential to normal growth and development, data are inadequate to establish that an exogenous source of carnitine is essential except in cases of genetic carnitine deficiency. In addition, data are incomplete on quantities of carnitine required for normal growth and development of infants and children and quantities necessary to maintain adequate carnitine nutrition of adults. For normal, healthy adults, there are no data that suggest beneficial nutritional effects from consumption of any form of carnitine.

Clinical studies indicate that orally administered L-carnitine may have beneficial effects in patients with carnitine deficiency states, renal disorders requiring hemodialysis, certain cardiovascular conditions, certain types of hypoglycemia, and type IV hyperlipidemia. In some patients, deficiency states and administration of DL-carnitine have been associated with myasthenia-like symptoms; alleviation of symptoms has been observed following administration of L-carnitine or withdrawal of DL-carnitine treatment. The use of exogenous L-carnitine for these conditions is currently investigational and should be considered separate and distinct from aspects of carnitine nutriture of normal individuals.

Orally administered D-carnitine and DL-carnitine are excreted by healthy adults, although the L-isomer in the racemic mixture may be retained differentially by tissues such as muscle. Animal studies consistently show that administration of the D-isomer, or the racemic mixture, is inferior to administration of L-carnitine in terms of affecting fatty acid oxidation or reducing experimentally induced cardiac ischemia. Further, intravenous,
intraperitoneal, or oral doses of DL-carnitine or D-carnitine induce several side effects including cardiac arrhythmias and muscle weakness; L-carnitine at equivalent doses does not induce such side effects. The inhibition of acyltransferases and reduction of serum L-carnitine following administration of D-carnitine or the racemic mixture suggests that the D-isomer may be a competitive inhibitor of L-carnitine in several metabolic processes. Further critical studies of differential nutritional, metabolic, and therapeutic effects of carnitine require both pure and isotope-labeled supplies of L-, D-, and DL-carnitine at reasonable costs.

The report provides additional conclusions and suggestions for future research consideration on topics discussed by the ad hoc review group.
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I. INTRODUCTION

A. BACKGROUND

Carnitine (γ-trimethylamino-β-hydroxybutyrate) is essential for the transport of long-chain fatty acids across mitochondrial inner membranes; however, knowledge of the scope of its metabolic functions in normal infants, children, and adults is incomplete. In the past several years, this quaternary amine has been shown to be of value in the treatment of certain dysfunctions of skeletal muscle, some cases of acute onset of hypoglycemia, some abnormalities associated with chronic hemodialysis, and possibly of ischemic heart disease and other cardiomyopathies. Recognition that certain debilitating metabolic disorders, such as the lipid storage myopathies, may be associated with disturbed carnitine metabolism led to clinical trials of its possible therapeutic value.

In recent years, the D-isomer, L-isomer, and racemic mixture of carnitine, as well as their acetyl and other acyl derivatives, and certain radiolabelled preparations, have been prepared on a limited basis by several commercial suppliers for investigational purposes. Data from several investigations suggest that metabolic responses to administration of the L- and D-isomers, the racemic mixture, or their respective derivatives may vary among subjects.

Because the Food and Drug Administration (FDA) is responsible for ensuring the safety and adequacy of the American food supply and for providing information on nutrition to consumers, the agency has an interest in the public availability of the several forms of carnitine and its derivatives. For this reason, FDA requested that the Life Sciences Research Office (LSRO) of the Federation of American Societies for Experimental Biology (FASEB) review available data on carnitine and obtain the opinions of knowledgeable investigators concerning health effects of orally ingested carnitine. LSRO convened an ad hoc review group on June 14-15, 1982, to discuss possible health effects of dietary carnitine.

B. SCOPE OF THE STUDY

The primary objective of this study is to identify the effects of oral ingestion of carnitine. Particular attention has been focused on the current state of knowledge regarding essentiality, differential metabolic effects of the optical isomers and the racemic mixture, and possible public health concerns arising from oral ingestion of quantities of carnitine that may be in excess of nutritional needs.
It was recognized that experimental data and clinical observations might be insufficient and that additional research might be necessary to document health effects. For example, data on absorption, metabolism, and excretion of dietary and endogenously synthesized carnitine in normal infants, children, and adults are limited. Much of the available data has resulted from animal or in vitro studies and most clinical data have been derived from studies of a few patients with metabolic disorders related to carnitine deficiencies, inborn errors of carnitine metabolism, or with patients on chronic hemodialysis. In addition, clinical trials have often utilized routes of administration other than oral ingestion.

The following and related questions were addressed in discussions of the ad hoc working group:

- What is known about qualitative and quantitative aspects of carnitine in the normal diet?

- What is known about bioavailability, absorption, and metabolism of carnitine and its derivatives in ostensibly healthy infants, children, and adults?

- Is carnitine an "essential" nutrient under certain circumstances? What are the relative roles of exogenous and endogenous carnitine in meeting metabolic needs?

- Is there experimental and/or clinical evidence that documents differential effects of carnitine isomers or derivatives on functions of cardiac, renal, hepatic, and other organ systems?

- What biochemical evidence is available on the specific differential effects of carnitine isomers or their precursors on acyl transfer and other intracellular metabolic processes?

- Are there any public health concerns related to consumption of carnitine isomers or derivatives in amounts in excess of those found in the normal diet?

- What further research needs can be identified as critical to understanding the possible effects of consumption of carnitine in excess of amounts known to be present in the animal and plant products of the normal diet?
II. SOURCES OF DIETARY CARNITINE

Because it functions as a carrier of acyl groups across mitochondrial membranes, carnitine and its derivatives are thought to be universally present in tissues where fatty acid oxidation occurs. Thus, while carnitine is present in a number of foods of plant and animal origin, foods derived from animal sources contain greater amounts of carnitine (Mitchell, 1978a). Borum (1983) stated that meat and dairy products are the major sources of carnitine in the U.S. dietary and that analysis of hospital diets suggested intakes of 2 to 100 mg/d. Many tissues synthesize L-carnitine, and this isomer is considered to be the natural form present in both plant- and animal-derived foods. Evidence for synthesis of the D-isomer in biological systems is lacking.

Mitchell (1978a), in a comprehensive review of carnitine, noted that data on carnitine content of various dietary items were limited. She collated data from a number of other investigators and provided values for "total assayed" carnitine ranging from 0 up to values of 0.13, 0.76, and 1.06 mg/100 g edible portion of leaf cabbage, cauliflower, peanut embryo, and wheat germ, respectively. In contrast, Mitchell (1978a) reported that values for various animal tissues used as food were significantly higher:

<table>
<thead>
<tr>
<th>Source</th>
<th>mg/100 g edible portion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eggs, chicken, whole raw</td>
<td>0</td>
</tr>
<tr>
<td>Beef liver, raw</td>
<td>2.60</td>
</tr>
<tr>
<td>Cow's milk, fluid</td>
<td>0.53-3.91</td>
</tr>
<tr>
<td>Chicken muscle, raw</td>
<td>4.55-9.10</td>
</tr>
<tr>
<td>Pork liver, raw</td>
<td>4.92</td>
</tr>
<tr>
<td>Cow's milk, nonfat, dry</td>
<td>15.0</td>
</tr>
<tr>
<td>Rabbit muscle, raw</td>
<td>21.0</td>
</tr>
<tr>
<td>Beef tenderloin, raw</td>
<td>59.8</td>
</tr>
<tr>
<td>Lamb skeletal muscle, raw</td>
<td>77.97</td>
</tr>
<tr>
<td>Sheep skeletal muscle, raw</td>
<td>206.28</td>
</tr>
</tbody>
</table>

According to Mitchell (1978a), carnitine in food is present as the free form as well as short- and long-chain fatty acid esters. Acetyl-carnitine and free carnitine appear to predominate in both plant and animal tissues used as foods. Hydrolysis of both short- and long-chain esters to free carnitine in the small intestine by pancreatic esterases is a logical expectation, but confirmatory data are incomplete.

Mitchell (1978a) enumerated several problems associated with determining actual carnitine content of foods as consumed. These included the extraction procedures and assay methods used, the chemical forms of the carnitine assayed, lack of confirmatory data, small number of replicate samples, and little knowledge of
the effects of food preparation on carnitine. She also noted that, because carnitine is water-soluble, cooking with moist heat would be expected to result in loss of carnitine. Dry heat preparation of foods in which protein denaturation occurs would be expected to destroy the mitochondrial transferase enzymes which utilize carnitine as a substrate, but effects on carnitine itself are unclear. While there are a number of laboratories that have assayed the carnitine content of various foods, few of these data have been published.

Marquis and Fritz (1964) developed an enzymatic assay method which provided greater potential for quantifying results than previously used bioassay or colorimetric methods. The enzymatic technique was modified by Cederblad and Lindstedt (1972) to a radioisotope assay capable of detecting quantities as low as 10 to 20 pnmol. Most investigators have used the radioisotope method of Cederblad and Lindstedt (1972) or modifications of this technique to assay for L-carnitine in biological samples. McGarry and Foster (1976) introduced further refinements for measuring free and total carnitine in plasma, but few studies report use of these methods with foods. Borum et al. (1979), using a slight modification of the Cederblad and Lindstedt procedure, reported that human milk contains 59 nmol/ml of carnitine; liquid formulas with soy protein isolate, casein, or egg white solids as protein sources contain from undetectable levels to 4 nmol/ml of carnitine; but liquid formulas with beef or milk as protein sources contain 50 to 656 nmol/ml carnitine.

Because biosynthesis of carnitine has been found in a variety of tissues from numerous species including humans, the importance of dietary carnitine has been a subject of some discussion. While it is generally agreed that nutritionally adequate amounts of carnitine can be obtained from animal-derived foods or synthesized in vivo by normal children and adults consuming normal diets, some evidence suggests that exogenous supplies of carnitine or its precursors can be limiting when endogenous synthesis is inadequate.

Accumulating evidence suggests that de novo synthesis in neonates is reduced (Borum, 1981). Schmidt-Sommerfeld and colleagues (1978, 1982) reported increases in adipocytic carnitine levels at the initiation of oral feedings in neonates and decreased carnitine intake by premature infants impairs fatty acid oxidation. In addition, some studies have reported low levels of plasma carnitine in persons with compromised nutritional status. For example, Khan and Bamji (1979) and Khan-Siddiqui and Bamji (1980) have demonstrated lower plasma carnitine levels in children and male adults with severe protein energy malnutrition. These authors suggested the reduced levels of dietary carnitine and precursor amino acids may be the cause of low plasma carnitine levels; however, they indicated that deficiencies in cofactors or enzymes involved in carnitine synthesis as well as altered patterns of utilization were also important considerations. Tanphaichitr
et al. (1980) reported that certain Thai adults whose diets contained low levels of animal-derived foods had compromised nutritional status as well as reduced plasma carnitine levels. Rudman et al. (1980) found that, among 271 patients with a diversity of disorders and diseases, only 20 of the 60 patients with alcoholic cirrhosis exhibited serum total carnitine and carnitine excretion levels that were below normal. The authors also reported that in this series of cirrhotic patients, biosynthesis of carnitine from diets containing adequate amounts of lysine and methionine as well as absorption of carnitine from oral or parenteral administration (500 μmol DL-carnitine/d) were impaired. On the other hand, Fuller and Hoppel (1983) have reported recently that total carnitine, free carnitine, short-chain acylcarnitines, and long-chain acylcarnitines in plasma of 20 patients with alcoholic cirrhosis were significantly elevated over those values observed in 30 normal subjects. They concluded that alcoholic cirrhosis is a hypercarnitinemic condition.

There is evidence that very low or undetectable quantities of carnitine are available in vegetarian diets (Borum, 1983), defined-formula diets (Borum et al., 1979), and solutions used for total parenteral nutrition (Hahn et al., 1982; Penn et al., 1981; Schmidt-Sommerfeld et al., 1982; Worthley et al., 1983). For example, Worthley et al. (1983) recently reported low plasma concentrations and reduced urinary excretion of carnitine in a patient who had been maintained on total parenteral nutrition for 1 year. The condition was corrected by intravenous administration of L-carnitine.

A further complication in understanding the role of exogenous (dietary) carnitine is the paucity of data on quantities required for normal growth and development as well as maintenance of a normal state of health. Cederblad and Lindstedt (1976) estimated rats weighing 160 g consumed 113 μg and synthesized 486 μg of carnitine daily, with a urinary loss of about 637 μg/d and a total body pool of 9.1 mg. Mitchell (1978a) concluded that these estimates may be misleading because of a discrepancy between amounts actually measured and kinetic data. Cederblad (1976) also reported that one adult female excreted more urinary carnitine than the amount fed, suggesting that the quantity of endogenously synthesized and dietary carnitine absorbed exceeded daily requirements. Studies on ketogenesis in experimental animal models provide some clarification of the interrelationships between ingested and endogenously synthesized carnitine.

In summary, a number of foods of both plant and animal origin contribute to dietary intake of carnitine; however, the latter are the most important sources. Data on the stereoisomeric form of carnitine in foods are limited. Most information suggests dietary carnitine of animal origin is the L-isomer; there are no reports of the natural occurrence of the D-isomer. Little is known about the stability of carnitine during food preparation.
III. METABOLIC FATE OF DIETARY CARNITINE

A considerable body of knowledge concerning carnitine metabolism has evolved in recent years from intensive study of its role as a carrier of fatty acid acyl groups between the cytoplasm and the mitochondria. For more detailed information, recent comprehensive reviews should be consulted (Bach, 1982; Borum, 1983; Bremer, 1977; Broquist and Borum, 1982; Frenkel and McGarry, 1980; Mitchell, 1978a,b,c; Pande and Parvin, 1976) and a recent symposium on biosynthesis and function (Bieber et al., 1982; Broquist, 1982; Henderson et al., 1982; Hoppel, 1982; Rebouche, 1982).

While L-carnitine is relatively abundant in skeletal muscle consumed as meat and occurs in other foods, a considerable, but as yet undetermined, portion of the metabolically active carnitine is synthesized endogenously by humans. The relative importance of ingested versus endogenously produced carnitine requires further elucidation.

In man, L-carnitine is synthesized predominantly by the liver and kidney from lysine, with methionine serving as the methyl donor. Other tissues, such as skeletal and cardiac muscle, are able to synthesize its precursor, γ-butyrobetaine, but are unable to hydroxylate this moiety. The pathway of carnitine biosynthesis in most higher animals involves a five step process (Figure 1); however, the regulation of this pathway is not fully elucidated.

Several investigators have established that the methylation of lysine and the conversion of ε-N-trimethyllysine to γ-butyrobetaine can occur in most tissues of both the rat and human (Broquist and Borum, 1982; Rebouche, 1982); however, the location of the final enzymatic step involving γ-butyrobetaine hydroxylase varies from one species to another. For example, Englard (1979) and Englard and Carnicero (1978) found no activity of this enzyme in the kidneys of rats, guinea pigs, or mice. The enzyme was present in rodent liver and also hamster and rabbit kidney. In cats, dogs, monkeys, and humans, γ-butyrobetaine hydroxylases are present in both kidney and liver (Rebouche, 1982). Once synthesized in the liver and kidney, carnitine is transported to skeletal and cardiac muscle via the circulatory system. Carnitine is actively accumulated by skeletal and cardiac myocytes. Carnitine is excreted as the free substance and esterified derivatives via the urine.

A. ABSORPTION

While the biosynthesis and metabolism of carnitine are relatively well understood, the process of absorption of dietary carnitine is less well known (Mitchell, 1978a). However, accumulating evidence indicates ingested carnitine is absorbed by the mucosal cells of the small intestine and enters the blood stream relatively slowly.
FIGURE 1. Biosynthesis of L-carnitine (from Broquist, 1982, with permission).
In the past several years, a number of clinical studies with patients exhibiting several conditions related to abnormal carnitine metabolism have shown that orally administered DL- or L-carnitine can raise blood carnitine levels (Anonymous, 1981). Karpati et al. (1975) found plasma carnitine levels rose from 314 µg/dl to 595 µg/dl in 3 hours after one 500 mg dose of DL-carnitine was given to an 11-year-old male. Angelini et al. (1976) reported that the serum carnitine level of one carnitine-deficient patient rose rapidly from about 2.5 to 12.5 µmol carnitine/100 ml serum within 30 minutes after ingestion of a 15 g oral dose of L-carnitine.

Gross and Henderson (1983a) have recently reported on absorption studies using both the isolated perfused rat intestine and direct injection of the rat duodenal lumen. Perfusion studies showed that less than 5% of a luminal dose of 2 to 6 nmols of either D- or L-carnitine was transported across the tissue to the perfusate in 30 minutes. The L-isomer was absorbed from the intestinal contents about twice as rapidly as the D-isomer. Similar results were obtained in live animal studies. After 60 minutes, 90% of the L-carnitine had accumulated in the intestinal tissue and was subsequently released to the circulation over the next several hours. On the other hand, only 80% of the D-carnitine had accumulated in 2 hours, but release to the circulation occurred similarly. Gross and Henderson (1983b) have shown that uptake of radiolabeled L-carnitine and L-acetylcarnitine is more rapid in the upper jejunal segment of the rat intestine than in other portions. In addition, they found acetylation occurring in the duodenum, jejunum, and ileum within 5 minutes of dosing. Uptake of both L- and D-isomers was a saturable process and high concentrations of D-carnitine, L-acetylcarnitine, and trimethylaminobutyrate inhibited L-carnitine uptake. Gross and Henderson (1983b) also reported differential retention of L-carnitine by muscle and greater excretion of D-carnitine unchanged in the urine 5 hours after administration. Urinary L-carnitine was approximately 40% acetylated, while urinary D-carnitine was completely unchanged. They concluded that the two isomers competed for tubular reabsorption in the isolated perfused kidney.

Recently, Rebouche (1983) has reported on growth of male weanling Sprague-Dawley rats fed carnitine-free diets with L-carnitine, D-carnitine, DL-carnitine, or γ-butyrobetaine added to the diet. While these studies focus on differential effects of the isomers and racemic mixture on growth of rats, (see p.28), the results show rats fed 0.05-1.0% L-carnitine- or 0.1-2.0% DL-carnitine-supplemented diets had increased L-carnitine concentrations in all tissues studied. In regard to effects of the dietary supplements, serum L-carnitine values after feeding for 32 days were:
<table>
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<tr>
<th>diet</th>
<th>animals (N)</th>
<th>serum nmol/ml*</th>
</tr>
</thead>
<tbody>
<tr>
<td>basal diet only</td>
<td>8</td>
<td>41.1 ± 6.14</td>
</tr>
<tr>
<td>+ 0.1% L-carnitine</td>
<td>6</td>
<td>102.0 ± 11.1a</td>
</tr>
<tr>
<td>+ 1.0% L-carnitine</td>
<td>8</td>
<td>138.0 ± 26.6a</td>
</tr>
<tr>
<td>+ 0.2% DL-carnitine</td>
<td>7</td>
<td>84.8 ± 13.7a</td>
</tr>
<tr>
<td>+ 2.0% DL-carnitine</td>
<td>5</td>
<td>97.2 ± 18.7a</td>
</tr>
<tr>
<td>+ 1.0% D-carnitine</td>
<td>4</td>
<td>31.4 ± 6.56b</td>
</tr>
<tr>
<td>+ 0.1% γ-butyrobetaine</td>
<td>3</td>
<td>91.1 ± 12.9a</td>
</tr>
<tr>
<td>+ 1.0% γ-butyrobetaine</td>
<td>4</td>
<td>86.8 ± 17.8a</td>
</tr>
</tbody>
</table>

* student's t-test used to determine statistical significance of test groups versus unsupplemented group; a = P<0.001, b = P<0.05

Rebouche (1983) also reported reduced carnitine biosynthesis in tissues of animals fed the supplements or injected with radiolabelled ε-N-trimethyllysine. Taken together, these results indicate increased absorption of L- and DL-carnitine, as well as γ-butyrobetaine from supplemented diets.

The studies of Gross and Henderson (1983a) and Rebouche (1983) suggest that dietary L-carnitine in the free state is readily absorbed by the rat and that absorption is probably associated with an active transmural absorption mechanism located primarily in the duodenal-jejunal portion of the small intestine. Similar results have been reported by Shaw et al. (1983) using everted rings of rat intestine. The studies of Gross and Henderson (1983a) and Rebouche (1983) also show differential absorption of the isomers or racemic mixture by the rat intestine. Clinical observations in patients given oral doses of L- or DL-carnitine are consistent with the occurrence of analogous absorptive processes in man, but confirmatory human studies with diets containing free carnitine or foods containing bound carnitine remain to be accomplished.

Determination of the rate or quantity of exogenous dietary carnitine absorbed by the intestinal mucosal cells may be complicated by the occurrence of biosynthetic activities. For example, Zaspel et al. (1980) have shown that the isolated perfused rat intestine can absorb ε-N-trimethyllysine and other potential carnitine precursors. In addition, they showed that the isolated or intact rat kidney absorbed and metabolized ε-N-trimethyllysine to γ-butyrobetaine more rapidly than the isolated or intact rat liver. However, Zaspel et al. (1980) also showed the isolated or intact rat liver absorbs γ-butyrobetaine more rapidly and, unlike the kidney, can convert it to L-carnitine.
Khan-Siddiqui and Bamji (1983) have shown that oral administration of 5 g doses of lysine to healthy adult males produced significant increases in plasma carnitine levels within 6 hours. Subjects receiving doses of tryptophan or threonine did not exhibit an analogous increase in serum carnitine. These authors suggested that carnitine may be synthesized from free trimethyllysine as well as from protein bound trimethyllysine.

Typical human diets can be expected to contain lysine, methionine, and the intermediates of the carnitine biosynthetic pathway. Because these precursor substances can be absorbed and because the several steps of endogenous synthesis occur in several tissues, use of specially labelled substances appears to be the most useful approach to the study of exogenous dietary sources of carnitine or its precursors.

B. TRANSPORT

It is generally assumed that absorbed carnitine is transported to various organ systems via the blood; however, the exact nature of the transport system requires further study. Bohmer et al. (1974) found the level of L-carnitine in serum of certain patients was reduced significantly with chronic hemodialysis. A number of studies have confirmed that hemodialysis does remove carnitine and acetylcarnitine from the circulating blood and reduce tissue levels of carnitine (Bartel et al., 1981; Guarnieri et al., 1980; Mingardi et al., 1980). Whether the effects of hemodialysis are on carrier proteins, on carnitine per se, or on both factors is not yet established.

In addition to these observations, several investigators have shown that membranes of certain tissues do bind and retain carnitine or carnitine derivatives (Bohmer and Molstad, 1980). In addition, binding by proteins has been identified in kidney cortex (Huth and Shug, 1980) and cells of rat brain cortex (Huth et al., 1981), as well as heart myocytes (Bohmer et al., 1977; Cantrell and Borum, 1982; Liedtke et al., 1981) and human epididymis (Borum et al., 1983). These several investigators have suggested that cells contain specific carnitine binding proteins.

Borum and York (1982) have reported that approximately 75% of the carnitine of whole blood is associated with the plasma and 25% with the red blood cells. They suggested that a carnitine-binding protein was associated with the membrane of the red blood cell. Their in vitro studies showed the binding is time-dependent, can be saturated, and is dependent on protein concentration. Borum and York (1982) also noted from inhibitor studies that sulfhydryl groups played an important role in the binding process.
The presence of membrane-bound proteins in several tissues including red blood cells that do bind carnitine suggests that a transport system in the blood and a cellular system for selective uptake is present in several tissues. In addition, altered carnitine transport has been observed in cardiomyopathic hamsters (York et al., 1983). There is little evidence to suggest that exogenous carnitine would be transported by different mechanisms from endogenously synthesized carnitine. There is some evidence that transport of D-carnitine and L-carnitine is quantitatively, and perhaps qualitatively, distinguishable (see p. 23).

C. METABOLISM

It is well established that carnitine is essential for the transport of long-chain fatty acids into the mitochondrial matrix where long-chain fatty acids are converted to acylcarnitine esters, transported across the mitochondrial membrane, and reconverted to carnitine and fatty acyl Coenzyme A derivatives (Figure 2). The long-chain acyl-CoA molecules undergo β-oxidation, liberating energy. Cardiac muscle cells depend upon this mechanism as an important source of metabolic energy (Neely and Morgan, 1974; Opie, 1979; Vary et al., 1981).

In addition to its role as a cofactor in the transfer of fatty acids from the cytosol to the mitochondrial matrix, carnitine has a role in ketogenesis (McGarry and Foster, 1980); and in thermogenesis in brown adipose tissue (Hahn and Skala, 1975). Carnitine appears to be involved in a number of other metabolic processes; however, additional investigations are required to confirm and extend these observations. These include:

1) the regulation of gluconeogenesis (Broquist and Borum, 1982; Slonim et al., 1981);
2) stimulation of fatty acid synthesis from malonyl CoA (Anonymous, 1980; Christophersen and Norseth, 1981);
3) stimulation of acetoacetate oxidation (Gravina and Gravina-Sanvitale, 1969);
4) hepatic catabolism of short- and medium-chain acyl-carnitine derivatives (Bieber et al., 1981, 1982);
5) transport of fatty acids into peroxisomes (Bieber et al., 1982);
6) branched chain amino acid catabolism (Bieber et al., 1982);
7) sperm maturation and motility (Brooks et al., 1974; Soufir et al., 1981);
FIGURE 2. The role of L-carnitine in the intramitochondrial transport of long-chain fatty acids (from Broquist, 1982, with permission).

The author identified the enzymes as 1) palmitoyl-CoA synthetase; 2) "outer" carnitine palmitoyltransferase; 3) carnitine-palmitoyl-carnitine translocase; 4) "inner" carnitine palmitoyltransferase.
8) reversal of immunosuppression associated with parenteral lipid nutrition (De Simone et al., 1982);

9) alteration of serum triglyceride and cholesterol metabolism (Bell, 1983; Guarnieri et al., 1980; Maebashi et al., 1978); and,

10) possible function as a neurotransmitter in the brain (Shug et al., 1979).

D. EXCRETION

Cederblad and Lindstedt (1971) concluded that animal studies suggested little or no degradation of carnitine prior to excretion and that urinary carnitine is the major excretory component. Khairallah and Wolf (1967) had earlier reported that rats excreted from 2 to 20% of a carboxy-labeled carnitine as β-methylcholine and 7% as $^{14}$CO$_2$. Evidence for the excretion of the former was not found by Cox and Hoppel (1973) and the role of the respiratory route remains to be confirmed.

Most data indicate that humans excrete free carnitine and acylcarnitines as the major catabolic products. Gruner et al. (1966) identified trimethylamine oxide in urine of subjects given oral carnitine. Maebashi et al. (1976) reported that increases in urinary carnitine were related to effects of fasting, and in normal subjects, adult males excreted more total carnitine than adult females. Frohlich et al. (1978) and Hoppel and Genuth (1980) reported both free carnitine and acylcarnitines are present in urine following periods of fasting of 24 to 36 hours. Valkner and Bieber (1982) observed that acetylcarnitine was the major acyl derivative found in urine; however, other acyl derivatives were also present. Hoppel and Genuth (1980) reported no significant differences in excretion patterns of normal and obese subjects. Excretion of urinary short-chain acylcarnitines rose as plasma levels of carnitine increased. In both normal and obese subjects, excretion of acylcarnitines increased more rapidly than that of free carnitine during the fasting period.
IV. EVIDENCE RELATED TO ESSENTIALITY

The oxidation of fatty acids as well as their transport across mitochondrial membranes requires \( L \)-carnitine as a component of the metabolic pathway. In this sense, \( L \)-carnitine may be considered as essential to normal growth and development; however, because it is synthesized endogenously, there is some question as to the absolute essentiality of an exogenous source of \( L \)-carnitine. However, there is evidence that suggests certain neonates and individuals with defects in carnitine metabolism benefit from administration of exogenous carnitine. This evidence is related to the evolving body of knowledge indicating that both fatty acid oxidation and ketogenesis are important to survival of newborn infants (Borum, 1981). In addition, Schmidt-Sommerfeld et al. (1982) and others (Penn et al., 1981) have shown that the decreased carnitine intakes of premature infants or neonates receiving total parenteral nutrition are associated with impaired fatty acid oxidation and ketogenesis. Because some neonates have limited ability to synthesize carnitine (Carlson, 1979), several investigators believe that carnitine from breast milk or other sources of nutrition is required by the newborn. For this reason, they have suggested that the possible role of carnitine as an essential nutrient for newborn infants be further investigated (Borum, 1981). The second body of evidence is related to the association of several genetic disorders with altered carnitine metabolism. These include systemic carnitine deficiency characterized by progressive skeletal myopathy and, frequently, cardiac enlargement (Chapoy et al., 1980; Engel and Angelini, 1973; Reboeche and Engel, 1981), as well as a familial endocardial fibroelastosis associated with an apparent defect in carnitine biosynthesis (Tripp et al., 1981).

Based upon a review of available information on carnitine content of infant diets and studies of endogenous synthesis in neonates, the Committee on Nutrition of the American Academy of Pediatrics (Carlson, 1979) concluded that premature infants fed intravenously and neonates fed casein or soy-based formulas received essentially no exogenous carnitine. Further, such infants have lower concentrations of plasma carnitine than infants fed human or bovine milk. They also noted that the ability of infants to synthesize \( L \)-carnitine appeared to be limited. The Committee recommended further study of the metabolism of exogenous carnitine in infants and the quantification of endogenous synthetic capability in the fetal and neonatal periods. The Committee also concluded that at that time (1979) evidence was insufficient to support the need for addition of carnitine to formulas and nutritional supplements for infants on a routine basis.

Novak et al. (1981a) reported that an exogenous source of \( L \)-carnitine facilitates the formation of acylcarnitines and fatty acid oxidation in normal infants fed a soy-based formula supplemented with 50 nmol/ml \( L \)-carnitine from the first or second day
after birth to 6 months of age. Novak et al. (1981b) have suggested that an optimal rate of fatty acid oxidation in neonates may depend on availability of exogenous carnitine. Schmidt-Sommerfeld et al. (1982) studied carnitine metabolism in 26 premature infants fed parenterally or fed human milk (or milk formula) with parenteral supplementation beginning between day 5 and day 27 after birth. In infants receiving parenteral lipid (Intralipid®) or either milk and supplemental parenteral lipid, plasma concentrations of acylcarnitines, β-hydroxybutyrate, free fatty acids and triglycerides increased, but free carnitine concentrations decreased after infusion. Total plasma carnitine values were relatively constant over the 4-hour lipid infusion period. Schmidt-Sommerfeld et al. (1982) interpreted their results as further evidence for increased fatty acid utilization in the neonate receiving lipid and suggested that absence or low levels of carnitine intake in premature infants may adversely affect the ability to oxidize fatty acids and produce ketone bodies. Further evidence of the importance of lipids as an energy source for neonates was reported recently by Cederblad et al. (1982). In a study of 13 normal, full-term male infants, carnitine excretion 3 days after delivery was similar to that at 6 days after delivery even though the quantity of breast milk ingested was greater on day 6. Newborns had a markedly increased acylation of excreted carnitine compared to that of healthy males aged 8 to 15 years. Penn et al. (1982) reported a decrease in plasma carnitine levels of 12 premature infants fed a carnitine-free parenteral formula during the first 5 days after birth. Urinary excretion of carnitine dropped to essentially zero during this period. These changes were not observed in eight similar infants fed a carnitine-containing formula. When oral feeding was initiated, plasma levels and urinary excretion of carnitine increased in the premature infants.

In a review of the evidence for a carnitine requirement of neonates, Borum (1981) summarized available experimental evidence as follows:

a) neonates have a critical need for carnitine because of the increased metabolic role of fatty acid oxidation;

b) plasma and tissue concentrations of carnitine of the neonate are low in comparison to those found in normal children and adults;

c) at birth, the ability to synthesize carnitine endogenously is not fully developed; and,

d) the lack of an exogenous supply of carnitine in the infant diet results in significant reduction in plasma concentrations of carnitine.
These data suggest that in the premature neonate, inadequate exogenous supplies or insufficient endogenous synthesis of carnitine may result in impaired fatty acid oxidation and ketogenesis, reduced thermogenesis, and decreased gluconeogenesis. When lipids are given to premature infants parenterally, the limited availability of carnitine may play a role in lipid toxicity. However, as pointed out by Borum (1981), the critical issue with respect to essentiality of dietary carnitine for neonates is related to the limited observations of altered physiology or pathology resulting from the deficiency state during the immediate postpartum period. Currently, available evidence is insufficient to establish the essentiality of exogenous carnitine in newborn infants.

Carnitine deficiency in children and adults is rare, and when diagnosed, is typically related to genetic deficiency states. During the past decade, relatively few individuals have been identified as carnitine-deficient (Borum, 1981). In most cases of systemic carnitine deficiency studied, the only defect identified is reduced capacity for renal absorption of carnitine; genetic defects in biosynthesis by the liver and kidney or transport into myocardial or skeletal muscle cells have not been observed as yet. However, Engel and Rebouche (1982), in a recent review of carnitine-deficiency syndromes, suggested that carnitine deficiency might also arise from impaired biosynthesis, impaired transport into cells, excessive cellular loss, excessive loss from body fluids, and/or excessive catabolic destruction.

Carnitine deficiency appears to be a spectrum of related syndromes involving progressive muscle weakness with lipid accumulation in Type I (slow) muscle fibers (Borum, 1981). The first diagnosed case, reported by Engel and Angelini in 1973, involved lipid storage myopathy. In the past decade a host of symptoms and signs have been reported including muscle carnitine deficiency, excess lipid deposition in muscle, various myocardopathies, peripheral neuropathy, myoglobinurina, and acute encephalopathy (Engel and Rebouche, 1982). Carnitine deficiency may be precipitated by other conditions, for example, in cirrhosis associated with cachexia (Rudman et al., 1977), in patients on prolonged hemodialysis (Bohmer et al., 1978), in propionicacidemia (Roe and Bohan, 1982), and in several other diseases (Engel and Rebouche, 1982).

Despite certain inadequacies, a tentative system of classifying carnitine deficiency states is in general use (Engel, 1980; McGarry and Foster, 1980). Primary syndromes are related to inborn errors of metabolism while secondary syndromes are acquired or associated with other congenital disorders. If the deficiency state involves only muscle cells, it is considered myopathic; where several tissues are involved, the deficiency state is considered systemic.
The actual incidence of primary and secondary as well as myopathic and systemic carnitine deficiency is unknown. One aspect of the problem is the capability of diagnostic centers to recognize and confirm the deficiency state (Borum, 1981), which suggests the condition may be more widespread than heretofore identified. However, agreement on diagnostic criteria has been lacking, and the criteria are currently under investigation and development. For systemic carnitine deficiency, the ad hoc review group suggested that plasma carnitine levels by themselves are not useful diagnostically. Low plasma carnitine levels are only suggestive or presumptive evidence of deficiency. Because patients with muscle carnitine deficiency may have normal serum carnitine concentrations, serum concentrations by themselves are not diagnostic. Furthermore, altered distribution between free and esterified forms of carnitine is not diagnostic. More discrete parameters suggested as diagnostic criteria include: lipid accumulation in Type I muscle fibers; decreased plasma carnitine concentrations after fasting; and, muscle cell concentrations of total or free carnitine about 25% or less of normal values.* Additional criteria suggested for further consideration included increased urinary excretion of $C_6-C_{10}$ dicarboxylic acids after fasting, relative levels of liver and kidney enzyme activities for the five enzymes known to be required for biosynthesis of carnitine, and red blood cell carnitine concentrations.

In most cases of systemic carnitine deficiency studied, the exact biochemical defect has not been established conclusively. Studies are continuing in several research laboratories; however, the limited number of patients and the complex nature of carnitine metabolism in several tissues are limiting factors in further elucidation of the precise basis or bases of systemic carnitine deficiency. The observation of acquired carnitine deficiency in several disease states, e.g., myopathies, and other disorders, e.g., chronic renal failure with hemodialysis, suggests that otherwise healthy individuals may either have subclinical deficiency states of possible genetic origin or may exhibit carnitine deficiency with metabolic stress or in certain disease states.

For example, Slonim et al. (1981) recently reported a case of an infant who had several bouts of severe hypoglycemia from age 4 weeks to 7.5 months. When the infant was fed a diet enriched with carnitine, clinical, biochemical, and histologic improvement was noted. More importantly, in one clinical trial

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* Rebouche and Engel (1983) have reported free and total carnitine levels in skeletal muscle for males as $18.0 \pm 8.1$ and $20.5 \pm 8.4$ nmol/mg of noncollagen protein (range 10-50, $N = 30$; range 12-22, $N = 30$, respectively) and for females as $17.3 \pm 5.3$ and $20.1 \pm 5.3$ nmol/mg of noncollagen protein (range 8-35, $N = 31$; range 11-39, $N = 31$, respectively), respectively.
when enrichment of the diet was withdrawn, plasma concentration
and urinary excretion rates fell to subnormal levels, and excre-
tion of dicarboxylic acids increased. Slonim et al. (1981) sug-
gested that these observations support the concept that dietary
carnitine may be essential if the ability to synthesize carnitine
is inadequate.

In summary, there are several unresolved questions related
to primary and secondary carnitine deficiency. When it does occur,
reasoned judgment and most experimental data indicate some defect
in endogenous synthesis or transport mechanisms. On the other
hand, limited clinical data indicate dietary carnitine can, in
some instances, overcome the deficit resulting from inadequate
endogenous synthesis. Thus, while carnitine may be considered
essential for a number of metabolic processes, the question of
essentiality of an exogenous supply must remain open until further
investigations have been completed.
V. DIFFERENTIAL EFFECTS OF CARNITINE ISOMERS

Major concerns of this report and the discussions of the ad hoc review group were the possible differential effects of the two isomers of carnitine as well as the metabolism of the racemic mixture. These issues are related to the universal occurrence of L-carnitine as a natural constituent of microbial, plant, and animal tissues and the virtual absence of the D-isomer in biological systems unless the synthetic substance has been introduced. Until recently, the introduction of the D-isomer alone, or as a component of the racemic mixture, was restricted largely to experimental animal studies or investigational therapeutic trials. More recently, DL-carnitine has been promoted as an over-the-counter preparation, raising additional questions about the metabolic consequences of oral ingestion of carnitine preparations containing the D-isomer. This issue has been exacerbated by the appearance of reports that DL-carnitine is effective in lowering serum triglycerides in hemodialysis patients (see p.29) and testimonials concerning increased capacity for physical performance. Thus, if used indiscriminately as an oral supplement to lower serum cholesterol and fatty acids or to influence physical performance, possible detrimental health effects in uninformed users might result.

A. ENZYMIC EFFECTS

Early studies of carnitine metabolism established that the two isomers of carnitine and their derivatives exert differential metabolic effects. Fritz and Schultz (1965) established that the L-isomers of carnitine and acetylcarnitine were substrates of carnitine acetyltransferase (EC 2.3.1.7) and that the D-isomers were competitive inhibitors. Fritz and Schultz (1965) and Fritz and Marquis (1965) also showed that D-acetylcarnitine inhibited fatty acid synthesis from glucose in adipose tissues, and that carnitine-induced stimulation of palmitate oxidation by rat heart mitochondrial preparations was competitively inhibited by D-palmitoylcarnitine. Most investigators now agree that D-carnitine is probably a competitive inhibitor of the L-isomer in all carnitine acyltransferase systems.

Holme et al. (1982) have reported that both carnitine isomers inhibit the γ-butyrobetaine hydroxylase reaction. This observation is significant in that the hydroxylase enzyme system is known to be stereospecific (England and Midelfort, 1978). Thus, the D-isomer, if present, could interfere with biosynthesis of L-carnitine from γ-butyrobetaine in the liver and kidney. Holme et al. (1982) noted that the myasthenia-like symptoms observed in hemodialysis patients administered DL-carnitine disappeared when L-carnitine was administered (Bazzato et al., 1981) and speculated that the symptoms might be related to the accumulation of the D-isomer and its effects on activity of γ-butyrobetaine
hydroxylase. There is no direct experimental evidence for this hypothesis; however, Welling et al. (1979) have shown 80% of a 40 mg/kg body weight intravenous dose of DL-carnitine was excreted unchanged in the urine within 24 hours. While rapid excretion of D-carnitine without metabolic breakdown or alteration appears to occur in adults who do not have renal disorders, additional studies are needed to confirm that D-carnitine is excreted in healthy subjects but accumulates in uremic patients producing adverse effects (Bazzato et al., 1981) (see also p.29).

B. INTERACTIONS WITH DRUGS

McGarry and Foster (1973) produced inhibition of hepatic ketogenesis in isolated perfused rat liver and in severely ketotic, alloxan-diabetic rats by means of D-decanoylcarnitine. Their studies were based on knowledge that the decanoyl-D-carnitine inhibited fatty acid oxidation by interference with long-chain acylcarnitine transferases. These results are consistent with the observations of competitive inhibition of transferase enzyme systems (Fritz and Schultz, 1965).

Alberts et al. (1978) showed that DL-carnitine suppressed or reduced the cardiotoxic effects of the antineoplastic agent, adriamycin. Maccari and Ramacci (1981), using isolated perfused hearts of male albino Wistar rats found the suppressive effects were related specifically to L-carnitine and that D-carnitine by itself did not protect against the cardiotoxicity of adriamycin.

Valproic acid is an anticonvulsant drug widely used in pediatric practice. An acute hepatic disease presenting as a Reye-like syndrome has been reported as a severe adverse effect. Böhles et al. (1982) recently reported decreased serum carnitine concentrations in a 3-year-old girl who developed acute liver disease after treatment with valproate for 6 months. Ohtani et al. (1982) studied 14 patients treated with a number of anticonvulsant drugs including valproic acid, 11 patients treated with several anticonvulsant drugs excluding valproic acid, and 27 age-matched control subjects. Plasma carnitine concentrations were lower and blood ammonia values were higher in patients treated with valproic acid than in untreated patients and control subjects. Both carnitine deficiency and hyperammonemia were corrected after oral administration of DL-carnitine (50 mg/kg/d) for 4 weeks.

C. ALTERED TISSUE AND CELLULAR TRANSPORT

The existence of an active transport system for uptake of L-carnitine is well established. Molstad et al. (1977) have shown that such a system is present in a cell line from human heart (CCL 27) and have used this model to study cellular efflux of L-carnitine as well (Molstad, 1980). Using tritium-labeled
L-carnitine to saturate binding sites in cultured cells, Molstad (1980) found that the rate constants for efflux of L-carnitine and D-carnitine at 100 μmol/l were essentially similar. Previous studies (Molstad et al., 1977) established that D-carnitine at 20 μmol/l reduced the uptake of L-carnitine to half the rate constant observed when L-carnitine was present alone. Molstad (1980) suggested that, while cellular mechanisms for influx and efflux are carrier-mediated, the difference in stereospecificity of the uptake and release mechanisms indicates that the two systems may not be identical. These in vitro studies suggest that while the D-isomer in the racemic mixture may interfere with cellular uptake of L-carnitine, both isomers would be transported out of cells at similar rates. Further evidence for competitive inhibition of L-carnitine uptake has been presented by Vary and Neely (1981). These investigators reported L-carnitine uptake in isolated perfused rat hearts involved both diffusion and a carrier-mediated transport system, and that D-carnitine competitively inhibited L-carnitine uptake (Ki=925 nmol D-carnitine/ml). On the other hand, Huth et al. (1979) showed that rat brain and kidney slices actively accumulated both D- and L-carnitine from the external medium. Rates of uptake of both isomers showed initial linearity followed by saturation with increasing concentrations of either isomer. While it is not possible to compare these two studies directly, the saturability observed by Huth et al. (1979) appears similar to the active mechanism rather than the diffusion process suggested by Vary and Neely (1981).

Huth and Shug (1980) reported subsequently that the carrier-mediated active transport mechanism in rat kidney cortex slices was associated with plasma membranes. While D-carnitine was also actively transported into cells by this carrier system, the carrier-mediated system favored L-carnitine. When D-carnitine was present, L-carnitine uptake was reduced to 64 ± 7% of the control uptake.

D. EFFECTS ON SKELETAL MUSCLE AND TISSUE CONTENT

Blum et al. (1971), in a comparative study of several forms of carnitine, choline, and acetylcholine, found differences in mean log dose response curves for the degree of contracture of isolated frog rectus abdominis muscles. In plots of log dose against contractile tension, L-carnitine was four times more potent than D-carnitine.

Maccari et al. (1982) reported that intraperitoneal administration of D-carnitine at 3 g/kg body weight daily for 4 days or 2 g/kg body weight each day for 30 days significantly reduced L-carnitine and L-acetyl carnitine content of heart, liver and tibial muscle of male albino Wistar rats. The extent of reduction of tissue concentrations was not reported. The investigators considered the effects of D-carnitine as a contraindication for use of DL-carnitine in treatment of diseases where carnitine deficiency impaired lipid metabolism.
E. EFFECTS ON CARDIAC MUSCLE

Results of studies on carnitine transport to specific tissues are consistent with the investigations of Welling et al. (1979) who followed the pharmacokinetics of L-carnitine after intravenous infusion of DL-carnitine (40 mg/kg body weight) into 14 male patients with coronary heart disease and one ostensibly normal male. L-carnitine distributed rapidly into the extracellular body water space and then more slowly into a second body compartment. Rates of perfusion suggested the latter was primarily fatty tissues and muscle. Welling et al. (1979) calculated from analysis of the urinary L-carnitine concentration that 80% of the DL-carnitine dose administered intravenously was eliminated in the urine. The investigators calculated that the administered dose of L-carnitine was about 6-10% of total body stores (estimated to be 256 mg/kg body weight).

In a separate study, these investigators (Thomsen et al., 1979) reported that these 14 patients, and others with coronary heart disease, showed improved cardiac pacing under physical stress after intravenous administration of 20 or 40 mg/kg body weight DL-carnitine. Taken together, these studies suggest that some of the D-carnitine may be eliminated unchanged in the urine following intravenous administration, and that at doses of 6-10% total body carnitine reserves, the possible detrimental effects of D-carnitine on metabolism of L-carnitine in ischemic human myocardium were insufficient to negate the potentially beneficial effects of L-carnitine.

These cardiac effects have been studied in other model systems. Liedtke et al. (1981) have shown that administration of DL-carnitine or L-carnitine to isolated, perfused, ischemic swine hearts partially restored the depleted tissue carnitine, decreased accumulation of long-chain acyl CoA, and improved mechanical functioning of the heart. L-carnitine (80 mg/ml) produced greater effects than DL-carnitine (170 mg/ml) when each was separately infused at 0.25 ml/min into the coronary arteries. In previous studies, Liedtke and Nellis (1979) had shown that perfusion of ischemic swine hearts with DL-carnitine (100 mg/kg) had no effect on tissue ATP levels, but did improve mechanical functioning when free fatty acids were also administered. In related studies, Suzuki et al. (1981) concluded that L-carnitine protected the ischemic myocardium of dogs by reducing the accumulation of long-chain acylcarnitine as well as long-chain acyl Coenzyme A. However, they found that perfusion of L-carnitine (100 mg/kg) prevented reduction in ATP content of ischemic myocardium while the DL-carnitine treatment was not effective. Suzuki et al. (1981) reported no ventricular arrhythmias or muscle weakness in dogs receiving doses of L-carnitine up to 100 mg/kg body weight, but made reference to such effects following DL-carnitine administration. In addition, they noted previous reports of myasthenia-like syndromes in hemodialysis patients (Bazzato et al., 1979).
and ventricular arrhythmias in dogs administered "high doses" of DL-carnitine intravenously (the calculated minimal dose inducing arrhythmias was 10.8 g over 15 minutes) (Brooks et al., 1977). Suzuki et al. (1981) stated that these side effects had not been observed in their experiments and concluded that L-carnitine had "more desirable effects" on ischemic myocardium than did DL-carnitine.

Paulson and Shug (1981) have shown that intraperitoneal administration of D-carnitine to starved and nonstarved male Sprague-Dawley rats resulted in tissue specific L-carnitine depletion. Administration of 200, 450, and 750 mg/day per 200-240 g rats for 4 days showed a dose-response relationship in that increased dose was related to progressively decreased total myocardial L-carnitine content. Paulson and Shug (1981) also reported that D-carnitine administration resulted in depletion of L-carnitine in skeletal muscle, heart, and kidney, but no changes in free, short-chain, or long-chain acylcarnitine content of liver, brain, or plasma were noted. In subsequent experiments with isolated perfused rat hearts, L-carnitine deficient hearts produced by D-carnitine administration failed to maintain coronary flow, aortic flow, and left ventricular pressure characteristic of hearts not treated with D-carnitine. They also reported accumulation of long-chain acylcarnitine in L-carnitine deficient hearts. Schaper et al. (1981) reported L-carnitine at 40 mg/kg body weight reduced infarct size in experimental occlusion studies (animal species not identified), but the racemic mixture, presumably at the same dosage level, had no effects on infarct size.

Garzya and Amico (1980) studied the effects of daily oral administration of 100 mg/kg body weight DL-carnitine or 50 mg/kg body weight L-carnitine in 20 patients with angina pectoris. The subjects used only nitroglycerine for 7 days, then isosorbide dinitrate (40 mg/d) for 14 days, then isosorbide dinitrate (40 mg/d) plus either DL-carnitine or L-carnitine from day 21 to day 81. The investigators followed the frequency of reported angina attacks and quantity of additional nitroglycerin used from day 21 to day 81. The 10 patients receiving L-carnitine and isosorbide dinitrate reported fewer angina attacks and used less nitroglycerine after day 30 than the 10 patients receiving DL-carnitine and isosorbide dinitrate. The authors pointed out their results were consistent with those of other investigators who had found clinical use of L-carnitine was preferred.

These studies may be viewed as clinical trials that extend knowledge derived from investigations of ischemic myocardial preparations in several animal models. While further double-blind studies of comparative effects of D-carnitine, L-carnitine, and the racemic mixture in animal models or patients with various forms of heart disease might provide further data on differential effects of the isomers, such studies in humans are constrained ethically by the several observations that, in most cases, L-carnitine is the experimental treatment that appears most efficacious.
Nevertheless, additional studies are needed to establish optimal dosages of L-carnitine, first in animal studies, and then in patients with various cardiac disorders where L-carnitine may prove to be therapeutically beneficial. Clearly such studies must be considered investigational drug trials rather than experimental efforts related to carnitine nutrition.

F. EFFECTS IN PATIENTS ON HEMODIALYSIS

Several studies have shown that hemodialysis removes circulating carnitine (Borum, 1983). In some patients, normal plasma carnitine levels are re-established after dialysis; in other patients, chronic dialysis leads to carnitine deficiency. Moorothy et al. (1983) have shown that skeletal muscle carnitine concentrations are reduced in hemodialysis patients. They postulated that hemodialysis extracts plasma carnitine resulting in unilateral transport of skeletal muscle carnitine stores to plasma. Several investigators have noted the decreased plasma concentrations of carnitine with hemodialysis and have administered carnitine via the dialysate or orally to treat the induced carnitine deficiency.

However, perhaps more important are observations that treatment with carnitine decreases the hypertriglyceridemia frequently found in hemodialysis patients (Lacour et al., 1980). As a result, a number of investigators have attempted to avoid a carnitine deficiency state and reduce serum triglycerides by adding L-carnitine to the dialysate or by administering L-carnitine or the racemic mixture intravenously before or after dialysis.

De Grandis et al. (1980) reported that 4 of 20 patients receiving 2.0 g of DL-carnitine three times each week after dialysis exhibited severe weakness of the biceps brachialis and extensor digitorum communis. The patients were dysphagic, and had difficulty chewing and swallowing. Neurophysiological tests confirmed the presence of neuromuscular transmission impairment. The four patients improved with bedrest and hemodialysis, but the reported symptoms of weakness and attendant neurophysiological disorders disappeared 4 to 5 days after DL-carnitine was withdrawn.

Bazzato et al. (1981) observed that 3 of 20 patients receiving 2.0 g of DL-carnitine intravenously three times per week after dialysis exhibited a myasthenia-like syndrome after 20 days of DL-carnitine administration. The patients had altered electromyograms and decreased skeletal muscle action potentials. Bazzato et al. (1981) also reported that symptoms and signs disappeared when DL-carnitine was withdrawn, but reported further that when the three patients were subsequently treated with L-carnitine for 45 days after a washout period, no muscle weakness was observed. These investigators concluded that the L-isomer was the drug of choice for dialysis patients when carnitine deficiency or Type IV hyperlipoproteinemia was evident.
Chan et al. (1982) noted that a myasthenia-like syndrome (inability to chew) was observed in 2 of 10 hypertriglyceridemic hemodialysis patients receiving 1.2 g of DL-carnitine daily. Withdrawal of carnitine treatment resulted in disappearance of the syndrome. More recently, Bellinghieri et al. (1983) and Vacha et al. (1983) have reported oral (2 g/d) and intravenous (20 mg/kg body weight) L-carnitine reduced hypertriglyceridemia and reduced muscle cramps and other symptoms in patients on hemodialysis; no myasthenic symptoms were noted by either group of investigators in the 43 patients studied. (See Chapter VI, p.29.)

G. ANIMAL FEEDING TRIALS

Until recently, the limited availability of D-carnitine precluded direct comparisons of the metabolic effect of oral ingestion of the stereoisomers of carnitine. In addition, lack of information on metabolism of ingested carnitine and its derivatives made such studies more difficult to interpret. However, two recent reports suggest differential metabolic fates of orally administered carnitine isomers.

Seim et al. (1980) have conducted a series of investigations on female Wistar rats and female AB Jena albino mice fed or subcutaneously administered D-carnitine, L-carnitine, their O-acyl derivatives, or DL-carnitine. In one study, two groups of five mice were given a total dose of either 3.3 g of D-carnitine or 3.5 g of L-carnitine (per os, in drinking water) for 31 days. No signs of intolerance were reported; however, only mice administered the D-isomer excreted the acetylolethylmethylammonium ion. Seim et al. (1980) stated that this specific derivative is formed spontaneously from dehydrocarnitine. Thus, Seim et al. (1980) reasoned that in the rodent, dehydrogenase enzymes can degrade D-carnitine, but do not metabolize L-carnitine. They also reported that the O-acyl derivatives were converted to the respective isomers prior to dehydrogenation (D-carnitine) or were involved in metabolic processes other than β-oxidation (L-carnitine). They suggested further that patients receiving treatment with DL-carnitine would excrete the acetylolethylmethylammonium derivative; however, no data beyond that from rodent studies were presented. The results of Seim et al. (1980) suggest that the D-isomer is metabolized in the rodent gastrointestinal tract, but by a β-oxidation pathway leading to excretion. Whether these observations reflect microbial activity or existence of an oxidative pathway in rodents is not clear. Whether such an analogous pathway exists in humans remains to be determined. In addition, these studies suggest possible differential metabolic handling of the two components of exogenous DL-carnitine.

Rebouche (1983) has recently reported on the effects of orally ingested carnitine isomers and the carnitine precursor, γ-butyrobetaine, in the rat. Groups of 4 to 10 male, weanling,
Sprague-Dawley rats were fed a carnitine-free diet (control) supplemented with L-carnitine, D-carnitine, DL-carnitine, or γ-butyrobetaine at 0.05 to 2.0% of the diet for 32 days. The rats fed diets supplemented with L-carnitine (0.10-1.0%) had increased L-carnitine concentrations in serum and in all tissues studied. Except for liver, tissue concentrations of L-carnitine in animals fed equivalent amounts of L-carnitine (as the racemic mixture) were increased over controls, but were consistently lower than concentrations found in tissues of rats fed L-carnitine alone. D-carnitine at 1% of the diet significantly reduced serum and heart L-carnitine concentrations from those observed in control animals. The effects of γ-butyrobetaine depended on the level of dietary supplementation. Dietary L-carnitine, D-carnitine, and γ-butyrobetaine (1%) reduced carnitine biosynthesis from ε-N-trimethyl-L-lysine in vivo. However, this decrease probably resulted from effects on γ-butyrobetaine transport into tissues, rather than effects on biosynthesis per se.

Rebouche (1983) also reported that mild diarrhea occurred in rats fed L-carnitine (0.5% of the diet) or DL-carnitine (1.0-2.0% of the diet) during the first 16 days of feeding and moderate diarrhea in rats fed 1.0% L-carnitine over the 32 days of the feeding trial. No other side effects were noted; however, in a separate study, D-carnitine had been shown to inhibit L-carnitine transport into rat renal brush border membrane vesicles (Rebouche and Mack, 1983).

Based on these comparative feeding trials, Rebouche (1983) concluded that orally administered L-carnitine is more effective than DL-carnitine in augmenting tissue stores of L-carnitine. Further, oral D-carnitine reduced tissue levels of L-carnitine suggesting competition for specific sites in cellular carnitine transport processes. While no gross pathological changes or altered growth rates were attributable to ingestion of diets containing 1.0% D-carnitine or 1.0% γ-butyrobetaine, he suggested that the interference with normal L-carnitine transport and storage might be indicative of metabolic alterations which could occur in rats fed D-carnitine or γ-butyrobetaine at these dietary levels for periods longer than 32 days.
VI. THERAPEUTIC USE OF CARNITINE

In addition to issues related to possible differential effects of the isomers and racemic mixture, questions have been raised about the effects of oral ingestion of amounts of carnitine in excess of nutritional needs. This issue is obscured by the absence of data that provide evidence of a quantifiable human requirement for dietary carnitine over and above that produced endogenously and by the paucity of data from investigations in which carnitine has been administered to normal subjects. The majority of studies reported in the literature deal with the therapeutic use of the L-isomer or the racemic mixture in limited numbers of patients.

Bazzato et al. (1979) studied the effects of 2.0 g of DL-carnitine administered three times per week to 15 patients undergoing hemodialysis for chronic renal failure. After 45 days (dose estimated to be equal to 0.9 g/d i.v.), they observed reduction in serum triglyceride levels, but in 3 of the 15 patients who had been on hemodialysis 8 to 10 years, a myasthenia-like syndrome of muscle weakness in the limb and jaw muscles developed between hemodialysis sessions. Because the carnitine was administered after each hemodialysis session and because the severity of symptoms was reduced by hemodialysis, Bazzatto et al. (1979) related the condition to the administered carnitine. They suggested the myasthenia might be related to accumulation of D-carnitine or a carnitine metabolite, or absence of its excretion in persons who were anuric. Subsequently, De Grandis et al. (1980) reported follow-up studies on these 15 plus 5 additional hemodialysis patients. Four of the 20 patients reported weakness, dysphasia, and difficulties in chewing and swallowing food after 30 days of receiving 2.0 g of DL-carnitine three times each week after hemodialysis. All clinical symptoms and neurophysiological signs of the myasthenia-like syndrome disappeared 4 to 5 days after termination of carnitine treatment. De Grandis et al. (1980) suggested their neurophysiological observations were consistent with some form of inhibition of choline uptake by motor neurons and subsequent reduction of acetylcholine at the neuronal junctions. In 1981, these investigators (Bazzato et al., 1981) found no development of symptoms in three patients when L-carnitine (rather than DL-carnitine) was administered intravenously. Since the same dosage regimen was used, the actual L-carnitine dosage administered (about 0.9 g/d) was twice that in the DL-carnitine studies.

Thomsen et al. (1979) reported that central venous infusion of 20 and 40 mg/kg body weight of DL-carnitine improved tolerance for exercise stress associated with increased heart rate and blood pressure. The intravenous doses given the 11 patients were approximately 1.0 to 2.0 g DL-carnitine per test period.
The authors reported the absence of any adverse reactions. In a related study, these investigators (Welling et al. 1979) reported central venous administration of 40 or 60 mg/kg of DL-carnitine to fifteen male subjects (age 44 to 66, mean 55 years) at doses equivalent to about 3.2 to 4.8 g. Based on pharmacokinetic data, the authors suggested the administered carnitine equilibrated with extracellular water space and more slowly with tissues. The administered dose was lost by urinary excretion and virtually all of the dose was accounted for in the urine within 12 hours after administration.

Guarnieri et al. (1980) investigated the effects of carnitine on hypertriglyceridemia in patients with chronic uremia. Eight patients received 0.5 g of carnitine (form not specified, but by inference presumed to be DL-carnitine) three times per week (equal to 0.2 g/d) for 8 weeks and subsequently received 1.0 g three times per week (equal to 0.4 g/d) for 6 weeks. Carnitine solutions were administered intravenously after hemodialysis. A second group of eight patients was treated similarly, but received a placebo infusion (not further identified). The investigators reported no side effects with the exception of "some euphoria" in carnitine-treated patients. This effect was not otherwise described or identified as a direct result of the treatment.

Data on observations from studies in which DL- or L-carnitine were administered orally are summarized in Tables 1 and 2. Except for the one normal subject included by Angelini et al. (1976) and the recent study of Rossi and Siliprandi (1982) on reduction of serum triglycerides and increase in high-density lipoprotein (HDL) levels over 10 to 15 weeks in two adult males, all subjects were being treated for the hypertriglyceridemia common to uremic patients after prolonged dialysis or for suspected carnitine deficiency.

In the approximately 130 patients receiving DL-carnitine in 13 studies since 1975 (Table 1), 6 investigators do not mention side effects or adverse health effects from doses of DL-carnitine ranging from 1.2 to 6.0 g/d. Five investigators note the absence of toxic or adverse effects and 2 report nausea and transient diarrhea with subsequent development of tolerance. On the other hand, Chan et al. (1980) reported development of severe neuromuscular transmission problems in 2 of 10 patients treated with 1.2 g/d DL-carnitine for hypertriglyceridemia as a complication of prolonged hemodialysis. They described the symptoms as analogous to those reported by Bazzato et al. (1979) who had administered similar dosage levels intravenously to treat the same condition in 3 of 15 patients.

In a recent review of patterns of response of dialysis patients to DL-carnitine treatment, Chan et al. (1982) called attention to the lack of predictability of serum triglyceride response. Those investigators noted that while Lacour et al.
<table>
<thead>
<tr>
<th>Dose/day</th>
<th>Period of Administration</th>
<th>Rationale for Administration</th>
<th>Subjects:</th>
</tr>
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<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>No.</td>
</tr>
<tr>
<td>1.2 g</td>
<td>10-14 weeks</td>
<td>hyperglycemic hemodialysis patients</td>
<td>10</td>
</tr>
<tr>
<td>2.0 g</td>
<td>5 months</td>
<td>systemic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>2.0 g</td>
<td>3 months</td>
<td>myopathic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>2.0 g</td>
<td>30 days</td>
<td>uremia</td>
<td>10</td>
</tr>
<tr>
<td>2.4 g</td>
<td>14 days</td>
<td>uremia</td>
<td>6</td>
</tr>
<tr>
<td>2.4 g</td>
<td>30 days</td>
<td>uremia</td>
<td>51</td>
</tr>
<tr>
<td>3.0 g</td>
<td>7 weeks</td>
<td>myopathic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>3.0 g</td>
<td>weekly, repeated</td>
<td>uremia</td>
<td>6</td>
</tr>
<tr>
<td>Dose/day</td>
<td>Period of Administration</td>
<td>Rationale for Administration</td>
<td>Subjects:</td>
</tr>
<tr>
<td>----------</td>
<td>-------------------------</td>
<td>-------------------------------</td>
<td>-----------</td>
</tr>
<tr>
<td>3.0 g</td>
<td>40 days</td>
<td>hyperlipoproteinemia (Types II &amp; IV)</td>
<td>26</td>
</tr>
<tr>
<td>4.0 g</td>
<td>3 months</td>
<td>myopathic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>4.0 g</td>
<td>~ 20 months</td>
<td>myopathic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>4.0 g</td>
<td>?</td>
<td>systemic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>4.0 g</td>
<td>18 months</td>
<td>systemic carnitine deficiency</td>
<td>1</td>
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<tr>
<td>5.5 g</td>
<td>at least 110 days</td>
<td>myopathic/ systemic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>6.0 g</td>
<td>21 days</td>
<td>systemic carnitine deficiency</td>
<td>1</td>
</tr>
<tr>
<td>6.0 g</td>
<td>28 days</td>
<td>uremia</td>
<td>4</td>
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### Table 2. Studies of Oral L-Carnitine Ingestion of Dosages of 1.0 g/d or Above

<table>
<thead>
<tr>
<th>Dose/day</th>
<th>Period of Administration</th>
<th>Rationale for Administration</th>
<th>Subjects:</th>
<th>Clinical Improvement</th>
<th>Side Effects</th>
<th>Remarks</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.0 g</td>
<td>10-15 weeks</td>
<td>normal</td>
<td>2 45, 50 yr M</td>
<td>Increased HDL; decreased serum TG</td>
<td>no</td>
<td>none reported by subjects or observed by investigators</td>
<td>Rossi and Sillprandl, 1982</td>
</tr>
<tr>
<td>1.0 g</td>
<td>12 days</td>
<td>methylmalonic-aciduria</td>
<td>1 8 yr M</td>
<td>yes</td>
<td>no</td>
<td>absence of adverse effects noted</td>
<td>Seccombe et al., 1982</td>
</tr>
<tr>
<td>1.5 g</td>
<td>4 months</td>
<td>hyperglycemic Remodialysis patients</td>
<td>10 ? ?</td>
<td>no</td>
<td>not reported</td>
<td>---</td>
<td>Auble et al., 1980</td>
</tr>
<tr>
<td>2.0 g</td>
<td>3 years ?</td>
<td>systemic carnitine deficiency</td>
<td>1 3.5 yr M</td>
<td>yes</td>
<td>yes</td>
<td>transient diarrhea</td>
<td>Chapoy et al., 1980</td>
</tr>
<tr>
<td>~ 2.0 g</td>
<td>?</td>
<td>uremia</td>
<td>10 ? ?</td>
<td>no</td>
<td>not reported</td>
<td>---</td>
<td>Demellia et al., 1981</td>
</tr>
<tr>
<td>3.0 g</td>
<td>2.5 months</td>
<td>familial endocardial fibroelastosis</td>
<td>1 11 mo F</td>
<td>yes</td>
<td>not clear</td>
<td>In discussion, the authors note mild diarrhea and body odor with high dose, unclear if patient exhibited these signs</td>
<td>Tripp et al., 1981</td>
</tr>
<tr>
<td>3.0 g</td>
<td>1 week</td>
<td>myopathic carnitine deficiency</td>
<td>1 10 yr F</td>
<td>yes</td>
<td>not reported</td>
<td>---</td>
<td>Angelini et al., 1976</td>
</tr>
</tbody>
</table>
Table 2. (continued)

<table>
<thead>
<tr>
<th>Dose/day</th>
<th>Period of Administration</th>
<th>Rationale for Administration</th>
<th>Subject</th>
<th>Clinical Improvement</th>
<th>Side Effects</th>
<th>Remarks</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>~ 5.0 g</td>
<td>8 weeks</td>
<td>myopathic carnitine deficiency</td>
<td>1 16 yr M unclear not clear</td>
<td>prednisone concurrently at 1 mg/kg body wt.</td>
<td>Griggs et al., 1981</td>
<td></td>
<td></td>
</tr>
<tr>
<td>15.0 g</td>
<td>1 dose</td>
<td>myopathic carnitine deficiency</td>
<td>1 10 yr F -- not reported</td>
<td>--</td>
<td>Angelini et al., 1976</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>normal</td>
<td>1 7 M</td>
<td>--</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
(1980) reported reduction in serum triglycerides in 38 treated patients, 13 patients (26%) exhibited no change or a paradoxical rise in serum triglycerides. They also pointed out that four of the uremic patients treated by Guarnieri et al. (1980) who responded paradoxically to DL-carnitine had serum carnitine levels that were similar to or above serum carnitine levels of the control subjects, even though Guarnieri et al. (1980) were unable to demonstrate any correlation between serum carnitine levels and triglyceride concentrations. Based on the observations of Aubia et al. (1980) that treatment of 10 hypertriglyceridemic dialysis patients with 0.5 g of L-carnitine three times per day did not lower serum triglyceride levels significantly in a 4-month clinical trial and their own data on paradoxical responses in patients, Chan and coworkers (1982) suggested that use of DL-carnitine for treatment of hyperlipidemia in hemodialysis patients was not indicated. They suggested further that some index of likely response such as pretreatment versus post-treatment serum carnitine levels should be established as a predictive measure.

In nine studies of 28 persons consuming L-carnitine, four make no mention of side effects of doses from 1.0 to 15.0 g/d, two state no occurrence of adverse effects, two are equivocal, and one (Chapoy et al., 1980) reports transient diarrhea in a 3.5-year-old male receiving 2.0 g/d over an extended period (see Table 2). It is clear from several studies (Bazzato et al., 1979; Guarnieri et al., 1980; Lacour et al., 1980; Pola et al., 1980; Rossi and Siliprandi, 1982) that oral or intravenous administration of DL- or L-carnitine alters serum lipid patterns in both normal and uremic subjects. Most investigators regard lipid-lowering effects as beneficial in ischemic heart disease (Thomsen et al., 1979) or hyperlipidemia and arteriosclerotic heart disease.

Studies to date have been conducted on a limited number of subjects but the absence of side effects of daily doses of 1.0 to 2.0 g L-carnitine and the possibility of beneficial effects on serum lipoprotein profiles suggest further investigational drug trials. However, these studies have been conducted only for periods of up to 3 to 4 months. The need for extended clinical trials in both normal adults and persons with the several types of hyperlipidemia is obvious.

The experience garnered from oral administration of DL-carnitine is almost exclusively related to treatment of suspected or confirmed primary or secondary carnitine deficiency or to treatment of hypertriglyceridemia in anuric hemodialysis patients. Data suggest that neuromuscular transmission effects occur in anuric patients, but this is far from clear. Prevailing opinion of responsible investigators suggests that L-carnitine, the natural isomer, is the preferred substance for further investigation despite the scattered clinical reports of clinical improvement of subjects receiving the DL-form. Further it should be noted that while the documentation of adverse health effects of daily dosages of 1.0 g or more of DL-carnitine is fragmented and incomplete, no controlled experimental trials have established any beneficial effects from regular oral ingestion in healthy individuals.
VII. CONCLUSIONS

Dietary Carnitine

- Metabolic requirements of higher animals including humans for carnitine are met both by consumption of foods containing bioavailable carnitine and endogenous synthesis in the liver and kidney.

- Available evidence indicates that L-carnitine is the isomeric form found in microorganisms, plants, and animal tissues. Foods of animal origin are the primary source of dietary carnitine.

Absorption and Transport

- When supplied in the diet, γ-butyrobetaine, L-carnitine, and DL-carnitine appear to be absorbed readily by an active transport system in the small intestine and by passive transluminal absorption in the small intestine.

- Reduction of blood carnitine levels during hemodialysis as well as identification of proteins that bind carnitine suggest that exogenous and endogenous carnitine may be transported to various tissues by specific carrier systems. The actual identity of the transport mechanisms remains to be determined.

- Cellular influx and efflux of carnitine are carrier-mediated processes in which the α-isomer is a competitive inhibitor of influx or efflux of the L-isomer.

Possible Requirement for Exogenous Carnitine

- The relative importance of exogenous versus endogenous carnitine in relation to normal growth and development requires further elucidation. While there is abundant evidence that L-carnitine is essential to normal growth and development of neonates, data are inadequate to establish that an exogenous source of carnitine is essential, except in cases of genetic carnitine deficiency which are thought to occur as a consequence of inborn errors of metabolism. In addition, data are incomplete on quantities of exogenous carnitine required for normal growth and development of infants and children and quantities necessary to maintain adequate carnitine nutriment of adults, if any.
For normal, healthy adults, there are no data that suggest beneficial nutritional effects from consumption of any form of carnitine. Indeed, some clinical evidence derived from patients with renal disease suggests adverse effects from consumption or administration of DL-carnitine.

Deficiency Syndromes

- Syndromes ranging from myopathic to systemic deficiency states have been associated with inadequate dietary intake of carnitine, inadequate endogenous production of carnitine from amino acid precursors, and possibly, increased excretion of acylcarnitines. Such disorders are clinically evident and diagnosis typically uncovers serum and other tissue concentrations of carnitine that are lower, or excretion that is greater, than those found in normal persons. However, the incidence of these disorders and the specific nature of the underlying genetic or metabolic derangements requires further study.

- Hemodialysis of patients with renal disease results in lower body stores of carnitine and chronic dialysis can lead to carnitine deficiency. In some patients, deficiency states and administration of DL-carnitine have been associated with myasthenia-like symptoms; alleviation of symptoms has been observed following administration of L-carnitine or withdrawal of DL-carnitine treatment.

- Clinical studies and related animal experiments indicate that orally administered L-carnitine as a therapeutic agent may have beneficial effects in patients with carnitine deficiency states, renal disorders requiring hemodialysis, certain cardiovascular conditions, certain types of hypoglycemia, and type IV hyperlipidemia. The use of exogenous L-carnitine for these conditions is currently investigational and should be considered separate and distinct from aspects of carnitine nutriment of normal infants, children, and adults.

Differential Effects of Carnitine Isomers

- While L-carnitine and the racemic mixture have been available commercially for several years, supplies of relatively pure L-carnitine and D-carnitine have been limited by the complexity and costs of manufacture. Further critical studies of differential nutritional, metabolic, and therapeutic effects of carnitine require both pure and isotope-labeled supplies of L-, D-, and DL-carnitine at reasonable costs.
Absorbed dietary L-carnitine is transported to liver and kidney as well as cardiac and skeletal muscle in healthy adults. Orally administered D-carnitine and DL-carnitine are excreted by healthy adults, although the L-isomer in the racemic mixture may be retained differentially by certain tissues such as muscle after active absorption and transport.

Animal studies consistently show that administration of the D-isomer, or the racemic mixture, is inferior to administration of L-carnitine in terms of affecting fatty acid oxidation or reducing experimentally induced cardiac ischemia. Further, intravenous, intraperitoneal, or oral doses of DL-carnitine or D-carnitine induce several side effects including cardiac arrhythmias and muscle weakness; L-carnitine at equivalent doses does not induce such side effects.

The D-isomer of carnitine is a weak competitive inhibitor of L-carnitine for some acyltransferase enzymes.

The occurrence of inhibition of acyltransferases and of reduced serum L-carnitine levels following administration of D-carnitine or the racemic mixture suggests that the D-isomer, when present, may be acting as a competitive inhibitor of L-carnitine in several metabolic processes.
VIII. SUGGESTIONS FOR FUTURE CONSIDERATION

Dietary Carnitine

- There is a need to expand and make readily available the existing data on carnitine content of plant and animal products used as foods, including components of traditional and vegetarian diets, defined formula diets, and products for total parenteral nutrition.

- The bioavailability of L-carnitine from selected plant and animal foods should be investigated. In addition, the effects of food processing on bioavailability of dietary carnitine require further study.

Absorption and Transport

- There is a need for additional investigation of dose responses to orally administered L-carnitine and its precursors to determine quantitative aspects of absorption, metabolism, and excretion.

- Limited studies in animals and humans suggest that both short- and long-chain esters of carnitine are hydrolyzed in the small intestine by pancreatic esterases, producing free carnitine for absorption and transport. These suggestions require further confirmation by means of critical studies of the absorption and bioavailability of exogenous long- and short-chain esters of L-carnitine in animal models and man.

- The occurrence and components of an active absorption system as well as the occurrence of passive absorption mechanisms for carnitine in the human small intestine require further study.

- While the major metabolic pathway for synthesis of L-carnitine and its acyl derivatives is known, additional investigations on mechanisms that regulate synthesis are needed. Similarly, further research on regulation of transport to and from tissues and cells, as well as transport among subcellular compartments, should be undertaken. For example, the exact nature of the mechanisms of carnitine transport requires further elucidation.
Possible Requirement for Exogenous Carnitine

- There is a need for carefully controlled balance studies that identify the quantitative aspects of nutritional requirements for exogenous sources of carnitine in normal healthy infants, children, and adults.

- There is a need to assess the progressive ability of premature and low-birth-weight neonates to synthesize carnitine.

- Preliminary observations suggest that cellular metabolism of carnitine may be affected by cofactors such as ascorbic acid, pyridoxal phosphate, niacin, and iron. The role of these and other cofactors in regulation of carnitine metabolism requires additional investigation.

Deficiency Syndromes

- Rapid and accurate techniques that do not require tissue biopsy are urgently needed for determination of carnitine status of both normal individuals and persons with possible carnitine deficiency syndromes.

- More extensive therapeutic use of oral L-carnitine must be based on expanded knowledge of the fate of exogenous carnitine and the factors controlling endogenous synthesis and degradation. In addition, such studies should include investigations on regulation of carnitine metabolism in normal infants, children, and adults.

- The beneficial effect of adding L-carnitine to the recirculated dialysate of patients with renal disorders requires further confirmation and critical evaluation.

- While not related to the role of dietary carnitine in healthy humans, orally administered L-carnitine has been shown to produce beneficial effects in animal models and patients with ischemic heart disease. There is a need to investigate critically the ranges of optimal and possibly toxic levels of L-carnitine in animal studies prior to more extensive use in patients with ischemic heart disease. Evolving knowledge of optimal dosages determined from carefully controlled animal studies should be used to establish dosages used in clinical trials.

- While animal models of ischemic heart disease are available, there is a need to identify animal models of carnitine deficiency states and renal disease.
Differential Effects of Carnitine Isomers

- There is a need to extend feeding trials of animals fed D-carnitine, DL-carnitine, or L-carnitine and its precursors at levels equivalent to concentrations found in human diets or at doses known to produce no adverse effects in humans to determine the metabolic effects of such diets on biosynthesis and metabolic activities of L-carnitine. Such studies should include extended feeding trials of 6 to 12 months duration.

- Additional studies of differential nutritional, metabolic, or therapeutic effects of carnitine require both pure and isotope-labeled supplies of L-, D-, and DL-carnitine at reasonable costs.
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