Effect of carnitine on lipid metabolism in the neonate. II. Carnitine addition to lipid infusion during prolonged total parenteral nutrition

The effect of carnitine administration on lipid metabolism and carnitine and acylcarnitine plasma values of newborn infants, given total parenteral nutrition for the first 7 days of life, was studied during a 4-hour infusion of Intralipid. An increase in plasma concentrations of total carnitine, free carnitine, and short-chain and long-chain acylcarnitine was found, but no significant change in triglycerides, free fatty acids, glycerol, or β-hydroxybutarate plasma values was noted, as compared with values obtained without carnitine administration. Moreover, the low free carnitine and short-chain and long-chain acylcarnitine plasma levels found in newborn infants after 7 days of total parenteral nutrition did not seem to impair the utilization of infused lipids. The results support the concept that the relation between the carnitine pool and lipid metabolism can be influenced by intravenous glucose infusion. Low carnitine plasma concentrations do not necessarily signify a depletion of body carnitine, and sufficient tissue carnitine concentrations can probably maintain good lipid utilization for an extended period. (J PEDIATR 104:436, 1984)

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UTILIZATION OF LIPID for energy requires the presence of carnitine for optimum transfer of long-chain fatty acids into the mitochondrial matrix, where β-oxidation takes place.[1] Newborn infants are unable to synthesize enough carnitine to maintain a blood concentration similar to that found in the fetus and in the adult, and dietary intake constitutes the main source of carnitine in the neonatal period.[2] In fact, newborn infants receiving total parenteral nutrition are at risk for developing low blood carnitine concentrations.[3,4] Lipids are now routinely used in parenteral nutrition. The newborn infant has a reduced capacity of utilizing the intravenously administered fat,[5] and the low carnitine blood concentrations may play an important role in this diminished ability to metabolize lipids.

Our study was designed to determine whether exogenous carnitine supplementation could be useful in maintaining optimal blood carnitine concentrations and thus in enhancing the utilization of the infused lipids.

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MATERIALS AND METHODS

Eleven newborn infants admitted to the Neonatal Intensive Care Unit of the University of Padua because of surgical problems were included in the study. Clinical diagnoses included esophageal atresia (two infants), necrotizing enterocolitis (three), omphalocele, duodenal atresia (three), septic paralytic ileus, and esophageal perforation.

TPN	Total parenteral nutrition

All infants received carnitine-free total parenteral nutrition exclusively throughout the first 7 days of life. The parenteral nutrition consisted of glucose, fat (Intralipid, AB Kabi, Stockholm), electrolytes, vitamins, and amino acids (Freamine, Baxter, Trieste, Italy).

The infants ranged in gestational age from 32 to 40 weeks (mean 36.4 weeks); their weights ranged from 1670 to 3140 gm (mean 2540 gm). All infants were appropriate for gestational age with respect to birth weight. After 7

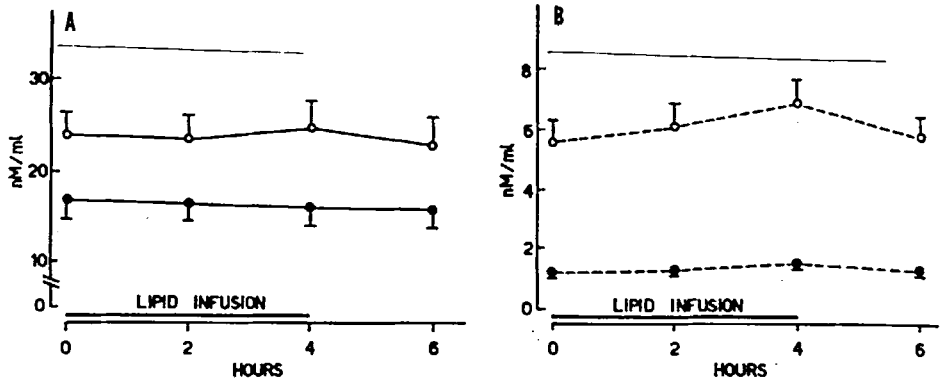

Fig. 1. Sequential changes in (A) total (o——o) and free (●——●) carnitine and (B) short-chain (o- - -o) and long-chain (●- - -●) acylcarnitine plasma levels during Intralipid tolerance test infusion (1 gm/kg body weight/4 hr) in newborn infants given TPN for 7 days. Values represent mean ± SEM.

days of total parenteral nutrition, a 10% Intralipid tolerance test, 1 gm/kg body weight over 4 hours, was given through a peripheral scalp vein using a constant infusion pump 12 hours after interruption of any intravenous lipid infusion.

Another Intralipid tolerance test, supplemented with L-carnitine (Sigma-Tau, Rome), 100 mg/kg body weight intravenously as a bolus before Intralipid infusion, followed by a continuous infusion of L-carnitine in a dose of 100 mg/kg body weight over 6 hours, was given 24 hours after the end of the first test. Blood samples were taken from a peripheral vein at 0, 2, 4, and 6 hours. A blood sample for basal plasma concentrations of carnitine and of short-chain and long-chain acylcarnitine was also drawn on the first day of life, before beginning TPN. Samples collected with heparin were immediately centrifuged at 3000 rpm; the resulting plasma was stored at −20° C until laboratory analysis. Total plasma concentrations of carnitine, free carnitine, and short-chain and long-chain acylcarnitines, were determined by a modification of the radioisotopic method of McGarry and Foster.[6] Tryglyceride and free glycerol were determined using the commercially available kit from Boehringer (Mannheim, F.R.G.). Free fatty acids were assayed with the enzymatic NEFA-C test (Wako Pure Chemical Industries, Osaka, Japan). β-Hydroxybutyrate was estimated according to the enzymatic fluorimetric method of Lloyd et al.[7] Values are given as mean ± SEM. The paired *t* test was used for statistical analysis. Written informed consent was obtained from the parents before undertaking the study.

RESULTS

After 7 days of total parenteral nutrition, the plasma values of total and free carnitine and of short-chain and long-chain acylcarnitine showed a significant decrease, compared with plasma concentrations on the first day of life (Table). After lipid infusion, a slight decrease in free carnitine and an increase in short-chain and long-chain acylcarnitine values were found (Fig. 1). The changes of carnitine and carnitine esters were small in comparison with those found by Schmidt-Sommerfeld et al.[8] using the same Intralipid tolerance test in newborn infants.

Table. Serum carnitine and acylcarnitine plasma levels in newborn infants given total parenteral nutrition for 7 days after birth

	Day 1	*Day 7*	*P*
Total carnitine (nM/ml)	37.2 ± 3.9	23.7 ± 2.5	<0.001
Free carnitine (nM/ml)	24.3 ± 2.8	16.6 ± 2.0	<0.005
Short-chain acylcarnitine (nM/ml)	10.8 ± 1.0	5.8 ± 0.7	<0.001
Long-chain acylcarnitine (nM/ml)	2.0 ± 0.2	1.1 ± 0.1	<0.05

Data expressed as mean ± SEM.

Carnitine supplementation of the Intralipid tolerance test produced a rapid increase of free carnitine plasma values (1108.1 ± 132.2 vs 16.4 ± 1.7 nM/ml in controls at the second hour of infusion; $P < 0.001$). The free carnitine plasma concentration was still significantly elevated at the end of the carnitine infusion (948.8 ± 142.2 nM/ml vs 15.7 ± 2.1 in controls, $P < 0.001$) (Fig. 2).

The changes in short-chain and long-chain acylcarnitine plasma values were parallel to those for free carnitine. After 2 hours, the plasma carnitine ester concentration was increased 10-fold, as compared with control values (long-chain acylcarnitine 10.7 ± 1.2 vs 1.2 ± 0.1 nM/ml, $P < 0.001$; short-chain 68 ± 13 vs 6 ± 0.3 nM/ml, $P < 0.001$). These values stayed elevated until the end of the carnitine infusion (long-chain acylcarnitine 9.1 ± 1.2

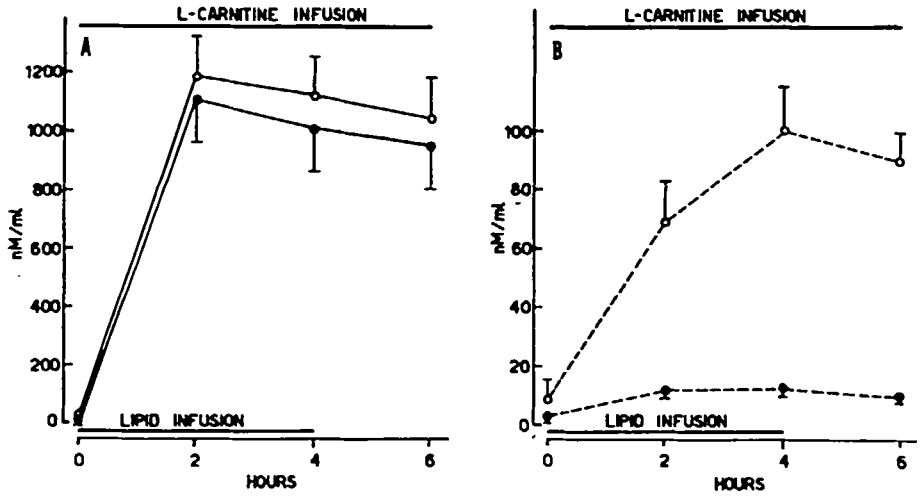

Fig. 2. Sequential changes in (A) total (o——o) and free (●——●) carnitine and (B) short-chain (o- - -o) and long-chain (●- - -●) acylcarnitine plasma levels after L-carnitine administration (100 mg/kg body weight as bolus and 100 mg/kg body weight as 6-hour continuous infusion) during 4-hour Intralipid tolerance test infusion in infants given parenteral alimentation for 7 days. Values represent mean ± SEM.

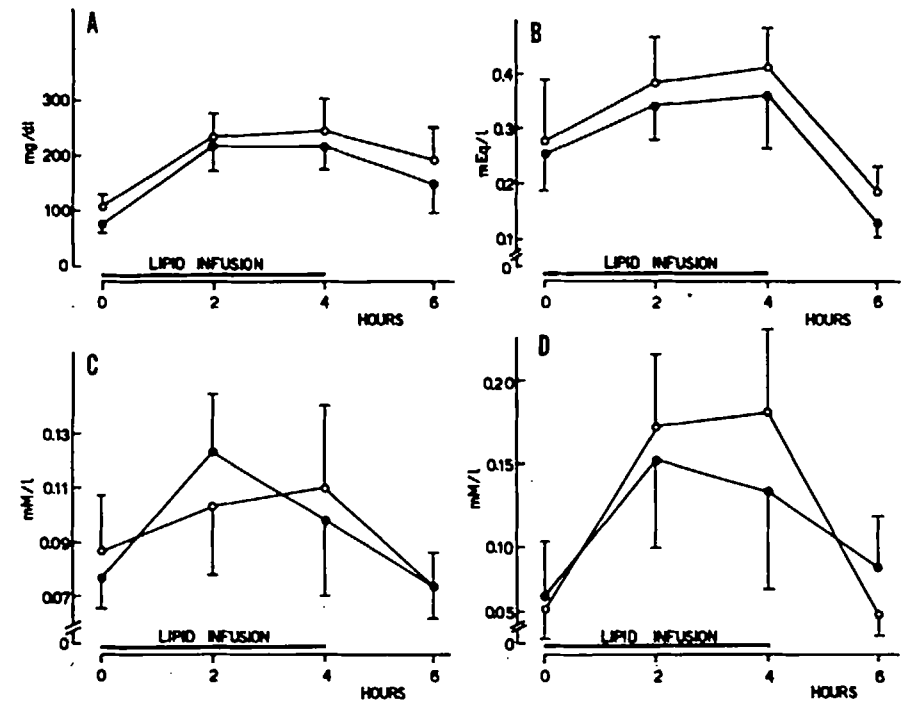

Fig. 3. Sequential changes in (A) triglyceride, (B) free fatty acids, (C) glycerol, and (D) β-hydroxybutyrate plasma levels during Intralipid tolerance test infusion with (o——o) and without (●——●) L-carnitine supplementation in infants given parenteral alimentation for 7 days. Values represent mean ± SEM.

vs 1.3 ± 0.1 nM/ml in controls, $P < 0.001$; short-chain 89.1 ± 9 vs 5.8 ± 0.7 nM/ml in controls, $P < 0.001$) (Fig. 2).

Plasma values of triglyceride, fatty acid, glycerol, and β-hydroxybutyrate increased during the Intralipid tolerance test (Fig. 3). The fatty acid, glycerol, and β-hydroxybutyrate plasma concentrations returned to preinfusion values 2 hours after the end of the lipid load. The plasma triglyceride concentrations were still elevated at this time, an indicator of a reduced removal rate of exogenous triglycerides from plasma.

Despite evidence that the exogenous carnitine was

acylated into the neonatal tissues and that large amounts of acylcarnitine were produced, no better utilization of infused lipids was noted (Fig. 3).

DISCUSSION

Our data confirm previous reports of the precariousness of carnitine availability to newborn infants dependent on long-term parenteral nutrition.[3,4] The low level of carnitine and carnitine esters found in neonates after 7 days of TPN raises the question as to whether these infants are capable of adequately utilizing a lipid infusion. Nevertheless, we found that newborn infants, despite low carnitine and acylcarnitine plasma levels, do not show significant differences in utilization of infused lipids before and after carnitine supplementation. This observation suggests that low carnitine plasma concentrations do not necessarily indicate depletion of body carnitine, and that lipid utilization can be maintained by a sufficient tissue carnitine concentration. We do not know, at present, how long these carnitine tissue stores can maintain good utilization of lipid infusions. Penn et al.[9] found low carnitine values in liver and heart, but not in skeletal muscle, only after very prolonged (>15 days) parenteral alimentation in newborn infants.

The formation of high levels of acylcarnitines after carnitine administration indicates that in infants the exogenous carnitine is taken up by the tissues (mostly the liver) and is integrated into the endogenous carnitine pool.[10] In our opinion, the large increase of these carnitine esters after carnitine infusion indicates a well-developed activity of the carnitine enzyme system, carnitine-palmitoyltransferase A and carnitine-acetyltransferase, in the neonatal period.

The plasma levels of long-chain and short-chain acylcarnitine (mostly acetylcarnitine) are considered important indicators of fatty acid turnover and ketogenesis.[11] A positive correlation has been shown to exist in vivo between serum β-hydroxybutyrate and acetylcarnitine levels.[8,12] Moreover, the addition of carnitine to the perfused liver system[13] and to isolated hepatocytes[14] increases ketone body production. In our earlier study,[15] we found increased ketone body production and lipolysis in newborn infants given parenteral alimentation supplemented with carnitine during an intravenous load of lipids in the first 48 hours of life; however, no such modification of lipid metabolism was found in this study of newborn infants after 7 days of total parenteral nutrition. These differences could be explained by different metabolic factors in the two groups of infants. In the first 48 hours of life, lipolysis and ketogenesis are enhanced; carnitine is known to play an important role under these conditions.[1,13] Moreover, no other intravenously administered solution was given during the 4-hour Intralipid infusion. On the contrary, the simultaneous infusion of glucose and amino acids with TPN during the lipid load in this second study probably reduces ketone body production. The glucose infusion increases serum insulin concentration, which in turn facilitates synthesis of triglycerides from the fatty acids in the liver. In our opinion, under this metabolic condition, ketogenesis is not enhanced by carnitine supplementation and the large amounts of acylcarnitines found in plasma after carnitine injection can be used for fatty acid esterification or energy production.

This concept can also explain the lower free carnitine decrease and acylcarnitine increase after Intralipid infusion found in our study, in comparison with the values obtained by Schmidt-Sommerfeld.[8] In fact, the newborn infants in Schmidt-Sommerfeld's study did not receive other solutions intravenously during the lipid load; consequently the β-hydroxybutyrate plasma values in these infants were found to be more elevated, in comparison with plasma values in our infants. The hypothesis that the regulation of ketogenesis may be dissociated from the regulation of β-oxidation has been suggested by Brass and Hoppel,[10,16] who found in rats infused with carnitine high levels of free carnitine and short-chain and long-chain acylcarnitine without changes in fatty acid and β-hydroxybutyrate concentrations. They support the concept that the carnitine pool reflects and results from the metabolic state of animals and that changes in the carnitine pool do not necessarily cause changes in the animal's metabolism.

In conclusion, it is our opinion that the question of the possible therapeutic benefits of supplementary carnitine in the infant receiving intravenously administered alimentation must be further investigated after more prolonged TPN and with long-term carnitine administration.

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