

Nutrient Interactions and Toxicity

Dietary L-Carnitine Improves Nitrogen Utilization in Growing Pigs Fed Low Energy, Fat-Containing Diets^{1,2}

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ABSTRACT Growing pigs ($n = 25$; 17.8 ± 0.1 kg) were used to study the effects of L-carnitine and protein intake on nitrogen (N) balance and body composition. Fat-supplemented (40 g soy oil/kg diet), corn-soybean meal basal diets containing low or high protein (136 or 180 g/diet) were formulated so that protein accretion would be limited by metabolizable energy (ME). Each basal diet was supplemented with 0 or 500 mg/kg L-carnitine and fed to pigs for 10 d in a nutrient balance trial. Final body composition was compared with weight and age-matched pigs measured on d 0 to calculate nutrient accretion rates. High protein feeding increased ($P < 0.01$) average daily gain (ADG) by 34%, as well as nitrogen digestibility (4.4%), retention (5.2%), urinary excretion (29%) and crude protein (CP) accretion (33%). Total-body carnitine accretion rate was 4.5 fold greater and total body carnitine concentration was almost 100% greater than in unsupplemented controls ($P < 0.01$). Irrespective of protein level, carnitine increased ADG (by 7.3%, $P < 0.10$) and CP accretion rate (9%, $P < 0.10$). Congruently, carnitine supplementation improved the efficiency of nitrogen retention ($P < 0.05$) and reduced urinary nitrogen excretion (14%, $P < 0.10$). Carcass fat content also was reduced in carnitine-supplemented pigs ($P < 0.10$). Collectively, these data support the hypothesis that carnitine can improve the efficiency of nitrogen utilization in 20-kg pigs fed energy-limited, fat-containing diets. We conclude that endogenous carnitine biosynthesis may be adequate to maintain sufficient tissue levels during growth, but that supplemental dietary carnitine (at 500 mg/kg) may be retained sufficiently so as to alter nutrient partitioning and thus body composition of 20-kg pigs. *J. Nutr.* 130: 1809–1814, 2000.

KEY WORDS: • pigs • carnitine • biosynthesis • nitrogen balance • body composition

Since the discovery of carnitine in the early 1900s, considerable research has elucidated its biochemical role in the transport of fatty acids across the inner mitochondrial membrane (see McGarry and Brown 1997 for review). Given that carnitine is a cosubstrate of carnitine palmitoyl-transferase, a pivotal regulatory enzyme in the pathway of fatty acid oxidation, carnitine status could conceivably affect utilization of fatty acids as metabolic fuel. This has led to interest and unsubstantiated claims among athletes and body-builders in the use of carnitine to enhance performance (Heinonen 1996). Similarly, interest in production agriculture has stemmed from the desire to partition nutrients away from fat accretion and toward muscle deposition. Several studies in a variety of species have reported improved nitrogen (N) balance, reduced body fat and/or increased protein accretion upon L-carnitine supplementa-

tion (Bohles et al. 1984b, Hongu and Sachan 2000, Ji et al. 1996, Penn et al. 1997, Rabie and Szilagyi 1998); however, the limited studies evaluating carnitine effects in growing pigs (Cho et al. 1999, Hoffman et al. 1993, Owen et al. 1996) have yielded inconsistent results.

Although biosynthesis of carnitine in the liver and kidney appears sufficient for the metabolic needs of mammalian adults (Rebouche and Seim 1998), dietary carnitine is necessary to maintain normal carnitine concentration in the newborn (Borum 1983). Indeed, the capacity of fatty acid oxidation in neonatal pigs (Coffey et al. 1991, Kempner and Odle 1993, 1995, Wolfe et al. 1978) depends on L-carnitine supply. However, it is not known how well growing mammals can synthesize carnitine de novo nor how dependent they are on carnitine supplied by the diet. In typical swine husbandry, pigs make a transition from a mixed-ingredient neonatal diet (containing various animal products) to a strict vegetarian diet (i.e., a corn-soy-based diet, devoid of animal products) at ~7–8 wk of age. Therefore, we chose to study pigs at this age on the basis of the supposition that removal of dietary carnitine sources (animal products) might occur while pigs were not fully competent with respect to de novo carnitine biosynthesis.

To test the hypothesis that dietary carnitine can alter

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nutrient partitioning in young growing pigs, we designed and fed basal diets [i.e., low metabolizable energy (ME)⁴/lysine with restricted intakes] such that N retention and protein accretion would respond to ME. Furthermore, diets were formulated to contain 7% fat on the supposition that supplemental carnitine would improve ME derived from fat (Fig. 1). Congruent with this hypothesis, the data reported herein will show that carnitine reduced urinary N excretion, increased protein accretion and reduced carcass fat.

MATERIALS AND METHODS

Animals and diets. All animal procedures were approved by the IACUC of North Carolina State University. Pigs ($n = 25$; 17.9 ± 0.1 kg; 54 ± 1 d of age) were used in five identical metabolic trials (5 pigs/trial) to study the interactive effects of L-carnitine and protein level on N and carnitine balance. Pigs (PIC genotype) were obtained from the Lake Wheeler Field Laboratory of North Carolina State University and fed corn-soybean meal diets (9 or 12 g lysine/kg diet) containing either 0 or 500 mg/kg added L-carnitine (2×2 factorial, randomized complete block design). Diets were formulated to contain 14.24 MJ ME/kg diet and 40 g/kg supplemental soy oil, and to exceed requirements for vitamins and minerals (NRC 1988) as shown in Table 1. The low protein diet was marginally adequate in protein, containing 0.63 g lysine/MJ ME. The high protein diet contained 0.84 g lysine/MJ ME. The diets were supplemented with crystalline amino acids to provide the same optimal ratios of essential amino acids (Chung and Baker 1992) to that of lysine.

Nitrogen balance. Pigs were placed into metabolism cages (0.77, 1.27 and 0.85 m in width, depth and height, respectively). After 5 d of adaptation to the 1.2% lysine diet without L-carnitine, pigs (~54 d of age) were allocated to experimental diets, and a 10-d nitrogen balance trial was conducted. The amount of feed offered was based on NRC (1987) formulas and previous measurements of ad libitum intakes of pigs during the adaptation period. To further ensure energy restriction, diets were offered at 85% of estimated ad libitum intakes. This also resulted in rapid and complete feed intake. All pigs were fed 400 g of diet twice (at 0800 and 2000 h) on d 1 of each metabolic trial, and the amount of feed per day was increased by 50 g every 2 d. Feed consumption was monitored along with total excretion of feces and urine. Urine was collected into a plastic bucket containing 30 mL

⁴ Abbreviations used: ADG, average daily gain; CP, crude protein; ME, metabolizable energy.

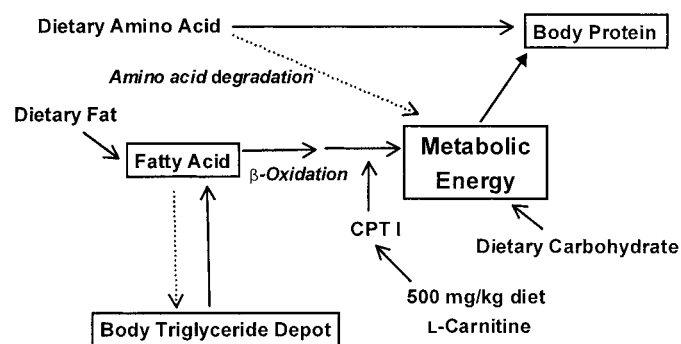


FIGURE 1 Illustration of the hypothesis that supplemental carnitine will improve nitrogen utilization for protein accretion by pigs fed a fat-containing basal diet but limited in metabolizable energy (ME). The diagram shows the primary metabolic fates of the central energy-yielding macronutrients. Bold arrows emphasize the predominant pathways, and dotted arrows indicate the hypothetically diminished pathways with L-carnitine supplementation. Dietary amino acids could provide needed metabolic energy in pigs fed limited ME, but L-carnitine may maximize dietary fat utilization via carnitine palmitoyltransferase I (CPT I), and thereby favor protein synthesis over amino acid degradation.

TABLE 1

Composition of low and high protein basal diets, as fed basis¹

| | Low | High |
|-------------------------------------|-------|-------|
| <i>g/kg diet</i> | | |
| Corn | 775.2 | 667.9 |
| Soybean meal | 136.3 | 244.7 |
| Soy oil | 40.0 | 40.0 |
| L-Lysine-HCl | 3.6 | 3.4 |
| DL-Methionine | 0.6 | 1.1 |
| L-Threonine | 0.5 | 0.8 |
| Dicalcium phosphate | 21.2 | 19.1 |
| Limestone | 5.7 | 6.3 |
| Salt | 3.5 | 3.5 |
| Vitamin-mineral premix ² | 3.3 | 3.3 |
| Antibiotic ³ | 10.0 | 10.0 |
| Calculated composition ⁴ | | |
| CP ⁵ | 136.0 | 180.0 |
| Fat ⁶ | 69.1 | 66.3 |
| Ca | 8.0 | 8.0 |
| P | 7.0 | 7.0 |
| Lysine | 9.0 | 12.0 |
| Methionine | 3.0 | 4.0 |
| Threonine | 5.9 | 7.8 |
| ME, MJ/kg diet | 14.24 | 14.24 |

¹ Each basal diet was fed with or without supplemental L-carnitine at 500 mg/kg diet. Low protein diets were formulated to contain 0.63 g lysine/MJ metabolizable energy (ME), and high protein diets were formulated to contain 0.84 g lysine/MJ ME. Analyzed L-carnitine (mg/kg) values (mean \pm SEM) in five trials were 3 ± 1 , 464 ± 10 , 6 ± 1 and 483 ± 14 for the low protein + 0 mg/kg L-carnitine diet (LP-0), low protein + 500 mg/kg (LP-500), high protein + 0 mg/kg (HP-0) and high protein + 500 mg/kg (HP-500) diets, respectively.

² Provided the following mg/kg of the complete diet: retinol, 2.2; cholecalciferol, 0.042; α -tocopherol, 22.1; menadione, 2.6; riboflavin, 5.8; niacin, 29; choline, 308; biotin, 0.08; pyridoxine, 1.45; folic acid, 1.13; D-pantothenic acid, 22; vitamin B-12, 0.029; Mn, 64; Fe, 104; Zn, 141; Cu, 25; I, 1.6; Se, 0.3.

³ Provided 55 mg of carbadox per kilogram of complete diet.

⁴ Calculated compositions were based on swine NRC (1988) values.

⁵ Analyzed crude protein (CP) (g/kg diet) values (mean \pm SEM) in five trials were 137.2 ± 1.2 , 137.4 ± 0.3 , 174.8 ± 1.6 and 177.2 ± 1.4 for the LP-0, LP-500, HP-0 and HP-500 diets, respectively.

⁶ Analyzed fat (g/kg diet) values in pooled five trials were 65.1, 66.6, 64.4 and 64.4 for the LP-0, LP-500, HP-0 and HP-500 diets, respectively.

of 6 mol/L HCl. A 50-mL aliquot of daily urine was stored at 4°C until the last day of the trial and was subsequently frozen at -20°C. Ferric oxide (0.25 g/100 g diet) was used as a marker to identify feces from the initial meal of the collection period. Total feces were collected daily, placed in aluminum trays and stored at 4°C. At the end of the test, total feces were desiccated in a 55°C air-forced drying oven. Fecal collections were subsampled and ground through a 1-mm screen, weighed and stored in plastic bags. Fecal and urine samples were analyzed for N by the micro-Kjeldahl procedure (AOAC 1990). Apparent nitrogen balance was computed as the difference between consumption and excretion.

Body composition analysis. At the beginning of each replicate of the metabolic trial, one pig was selected randomly and killed for measurement of initial empty body composition [percentage of protein, lipid and ash; AOAC (1990)]. At the end of each metabolic trial, (pig age = 64 d), all pigs were killed by electrocution. Except for the contents of the gastrointestinal tract and bladder, and small tissue biopsies taken, the entire carcass was ground once through a 15-mm diameter plate, twice through a 9-mm plate and then twice through a 3-mm plate. Subsamples of the final mixture were taken and stored at -20°C until they were analyzed for dry matter, ash, crude protein (micro-Kjeldahl), crude fat, total carnitine and energy content.

TABLE 2

Effects of L-carnitine and protein level on daily gain and nitrogen balance of 20-kg pigs¹

| | Low protein level | | High protein level | | SEM |
|---|-------------------------|-------|--------------------|-------|------|
| | L-Carnitine, mg/kg diet | | | | |
| | 0 | 500 | 0 | 500 | |
| Daily gain, g ^{ab} | 350 | 366 | 458 | 501 | 15 |
| Gain/Feed, g/kg diet ^{ab} | 412 | 431 | 539 | 589 | 18 |
| Nitrogen balance | | | | | |
| N intake, g/d ^a | 18.66 | 18.69 | 23.77 | 24.10 | 0.16 |
| N fecal excretion, g/d | 3.11 | 3.27 | 3.24 | 3.20 | 0.15 |
| N urine excretion, g/d ^{ab} | 2.90 | 2.59 | 3.88 | 3.22 | 0.23 |
| N retained/N intake, % ^a | 67.78 | 68.66 | 70.09 | 73.38 | 1.22 |
| N digestibility, % ^a | 83.31 | 82.52 | 86.39 | 86.71 | 0.80 |
| Biological value, ² % ^d | 81.34 | 83.22 | 81.14 | 84.63 | 1.21 |

¹ Values are means, $n = 5$ pigs/treatment.

² N Retained/N absorbed $\times 100$.

a,e Effect of protein level ($P < 0.01$, $P < 0.10$, respectively).

b,c,d Effect of L-carnitine ($P < 0.10$, $P < 0.01$, $P < 0.05$, respectively).

Chemical analysis was conducted on each sample in duplicate. From the chemical analysis, the total amounts of protein, lipid, ash and moisture were determined for each pig on an empty body weight basis. Thus, the initial carcass composition was subtracted from the composition determined at d 64 of age. Daily protein and lipid accretion rates were calculated as the difference between final (d 64 of age) and initial (d 54 of age) composition, divided by 10 d.

Blood sampling. Blood samples were obtained in heparinized tubes by vena cava puncture, 2–3 h after feeding on the last morning of each balance trial. Blood samples were centrifuged at $2300 \times g$ for 25 min within 1 h of collection. Plasma samples were stored at -20°C and analyzed for free carnitine by a radioenzymatic method.

Carnitine analysis. [$1-^{14}\text{C}$]acetyl-CoA was purchased from American Radiolabeled Chemicals (St. Louis, MO). Scintillation fluid (Scintisafe) and Resin (AG 1×8 , 100–200, chloride form) were obtained from Fisher Scientific (Fair Lawn, NJ) and Bio-Rad Laboratories (Richmond, CA), respectively. Acetyl-CoA, carnitine acetyltransferase (EC 2.3.1.7) and other chemicals were purchased from Sigma Chemical (St. Louis, MO). All samples were prepared by the procedure modified from Bhuiyan et al. (1992). Weighed sub-samples of diet, and ground body tissue were homogenized in appropriate volumes of water using a PowerGen Homogenizer (Model 700, Fisher Scientific, Atlanta, GA; 4×10 s at 30,000 rpm). Those mixtures and urine were alkalinized with KOH and incubated for 1 h at 60°C before treatment with HClO_4 . Then, the supernatant was neutralized for total carnitine assay. Free plasma carnitine and total carnitine of the samples described above were assayed by the enzymatic radioisotope method of McGarry and Foster (1976), modified as described by Bhuiyan et al. (1992). Carnitine concentrations of samples were corrected by blanks without acetylcarnitine transferase that were measured for each sample type and determined by using a standard concentration curve. The average recovery of free carnitine and total carnitine was 99 and 90%, respectively (data not shown).

Statistical analysis. All data were analyzed as a randomized complete block design with a 2×2 factorial (L-carnitine \times protein level) arrangement of treatments using the General Linear Models procedure of SAS (1989). The statistical model included 4 df for replicate, and 1 df each for L-carnitine, protein level and the interaction between L-carnitine and protein level. In addition, daily N excretion data were analyzed as above with an additional split-plot in time (Steel et al. 1997). This daily-excretion statistical model included 12 df for whole-plot error (trial \times protein level \times L-carnitine) and 4 df each for protein \times time and carnitine \times time interactions. The experimental unit was the individual pig in all statistical analyses. The relationships between tissue carnitine and nutrient accretion were determined by regression analysis. Significant differences and relationships were accepted at $P < 0.1$.

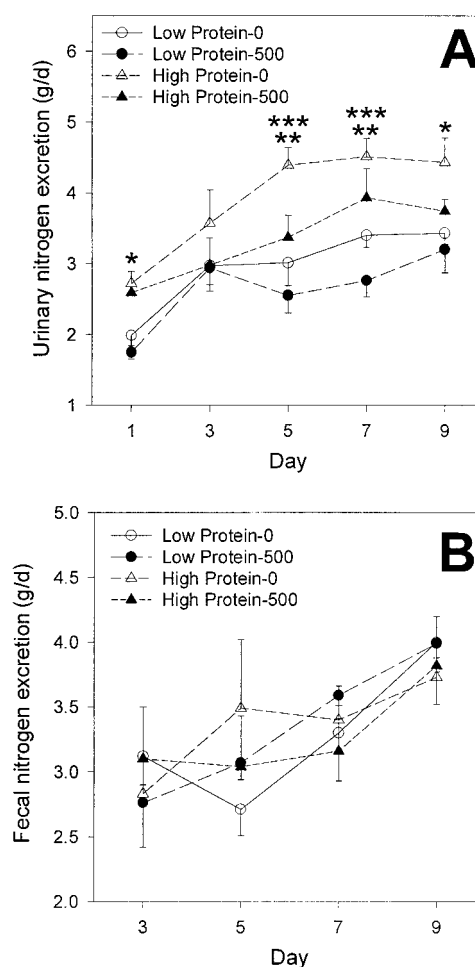


FIGURE 2 Effect of dietary L-carnitine and protein levels on daily nitrogen excretion of 20-kg pigs. Open and closed symbols refer to 0 and 500 mg/kg dietary carnitine, respectively. Each value represents the mean \pm SEM, $n = 5$ pigs. (A) Urinary excretion. Protein level main effect ($P < 0.01$); L-carnitine main effect ($P < 0.10$); protein level \times L-carnitine interaction ($P < 0.10$); protein level \times time interaction ($P < 0.05$); time main effect ($P < 0.01$); protein level effect within each respective day ($*P < 0.05$, $**P < 0.01$); $***$ L-carnitine effect within each respective day ($P < 0.05$). (B) Fecal excretion.

TABLE 3

Effects of L-carnitine and protein level on body composition and nutrient accretion rates of 20-kg pigs¹

| | Low protein level | | High protein level | | SEM |
|--|-------------------------|--------|--------------------|--------|-------|
| | L-Carnitine, mg/kg diet | | | | |
| | 0 | 500 | 0 | 500 | |
| Body composition, ² g/100 g | | | | | |
| Moisture | 67.70 | 67.59 | 67.94 | 68.13 | 0.36 |
| Protein | 16.36 | 16.52 | 16.51 | 16.63 | 0.14 |
| Fat ^a | 10.84 | 10.52 | 10.69 | 10.03 | 0.27 |
| Ash ^b | 3.36 | 3.29 | 3.17 | 3.16 | 0.06 |
| Carnitine, nmol/g ^c | 391 | 751 | 415 | 766 | 25 |
| Accretion rates, ³ g/d | | | | | |
| Moisture ^d | 210.85 | 206.72 | 272.56 | 302.89 | 13.16 |
| Protein ^d | 58.70 | 61.52 | 75.61 | 84.68 | 3.14 |
| Fat | 51.41 | 45.53 | 57.15 | 47.75 | 5.81 |
| Ash | 15.98 | 14.47 | 14.86 | 15.90 | 1.39 |
| Carnitine, μ mol/d ^c | 124 | 867 | 212 | 989 | 56 |

¹ Values are means, $n = 5$ pigs/treatment. A total of 25 pigs in 5 identical trials, 4 pigs/trial were used for final body composition, and 1 pig/trial was used to calculate the initial empty body composition of treatment pigs for nutrient accretion rates.

² Whole-body composition except for the contents of the gastrointestinal tract and urine contained within the bladder (i.e., empty body weight).

³ Initial body composition of the five littermate pigs was as follows: $68.85 \pm 0.33\%$ for moisture, $16.17 \pm 0.29\%$ for CP, $9.95 \pm 0.52\%$ for fat, $3.08 \pm 0.12\%$ for ash and 394 ± 25 nmol/g for carnitine.

a,c Effect of L-carnitine ($P < 0.10$, $P < 0.01$, respectively).

b,d Effect of protein level ($P < 0.05$, $P < 0.01$, respectively).

RESULTS

Growth performance and nitrogen balance. High protein feeding increased average daily gain (ADG) ($P < 0.01$, Table 2) by 34%, and L-carnitine supplementation increased ADG by 7.3% ($P < 0.10$). High protein feeding improved N digestibility ($P < 0.01$) by 4.4% and N retention ($P < 0.01$) by 5.2%, but increased urinary N excretion by 29% ($P < 0.01$). Carnitine reduced urinary N excretion by 14% ($P < 0.10$) and improved the biological value (defined as the percentage of absorbed N retained in the body) by 3.3% ($P < 0.05$). No interactions were detected between L-carnitine and protein level ($P > 0.10$).

Daily carnitine and nitrogen excretions. Carnitine supplementation increased daily urinary carnitine excretion ($P < 0.01$) linearly over time, but neither carnitine nor high protein feeding had any effect on daily fecal carnitine excretion (data not shown). High protein feeding had negligible effects on urinary or fecal daily carnitine excretions. Dietary L-carnitine reduced daily urinary nitrogen excretion after 3 d of supplementation ($P < 0.01$, Fig. 2A). High protein feeding maintained higher daily urinary N excretion for the entire experimental period ($P < 0.05$). Neither L-carnitine nor high protein feeding affected daily fecal N excretion ($P > 0.10$, Fig. 2B).

Body composition and nutrient accretion rates. Body composition data showed that the percentage of fat in the carcass decreased when L-carnitine was supplemented ($P < 0.10$), but the proportions of crude protein (CP) and moisture were not affected by either high protein feeding or L-carnitine ($P > 0.10$, Table 3). Body ash percentage was reduced by high protein feeding ($P < 0.05$). Carnitine concentration in carnitine-supplemented pigs was almost 100% higher than in control pigs ($P < 0.01$). High protein feeding increased CP accretion by 33% ($P < 0.01$) and moisture accretion by 38% ($P < 0.01$), but did not affect ($P > 0.10$) fat or ash accretion. Carnitine supplementation increased the CP accretion rate by $\sim 10\%$ ($P < 0.10$), and increased the carnitine

accretion rate 4.5-fold ($P < 0.01$). There were no interactions between L-carnitine and protein level ($P > 0.10$).

Correlation analysis. There were positive correlations between plasma free carnitine, body carnitine (Fig. 3A, $r^2 = 0.726$, $P < 0.001$) and urinary carnitine excretion (not shown). Deviate relationships between plasma free carnitine and protein accretion rate for the two protein levels are shown in Figure 3B. Protein accretion rate was positively correlated with plasma free carnitine in pigs fed high protein diets ($r^2 = 0.500$, $P < 0.023$), but not in pigs fed low protein diets ($r^2 = 0.001$, $P = 0.92$). Muscle carnitine concentration was positively correlated with protein accretion rate only in pigs fed high protein diets ($r^2 = 0.457$, $P < 0.032$, Fig. 3C).

DISCUSSION

Growth performance and nitrogen utilization. Studies evaluating L-carnitine effects on growth performance in young pigs (Cho et al. 1999, Hoffman et al. 1993, Owen et al. 1996) have reported variable findings. Cho et al. (1999) observed no appreciable improvement in the performance of 21-d-old pigs when 1000 mg/kg carnitine was supplemented into diets containing 17% dried skim milk. Owen et al. (1996) reported that supplementation of 500 mg/kg L-carnitine improved feed efficiency by 9% in pigs from d 36 to 57 of age when pigs were fed a corn-soybean meal-dry whey diet containing 5% soy oil. This improvement stemmed from reduced feed intake rather than increased ADG, suggesting that pigs fed L-carnitine may improve energy utilization from soy oil, but that control pigs fed no L-carnitine may satisfy their energy requirement by increasing feed intake. Hoffman et al. (1993) reported that supplemental L-carnitine or soybean oil did not affect ADG, energy or nitrogen utilization in young pigs. Because pigs in these experiments were allowed ad libitum access to feed, energy status was likely high enough to maintain ADG regardless of dietary carnitine. On the other hand, we suggest that energy limitation in our study (i.e., low ME/lysine treatment and 85% ad libitum consumption) accentuated energy utilization from

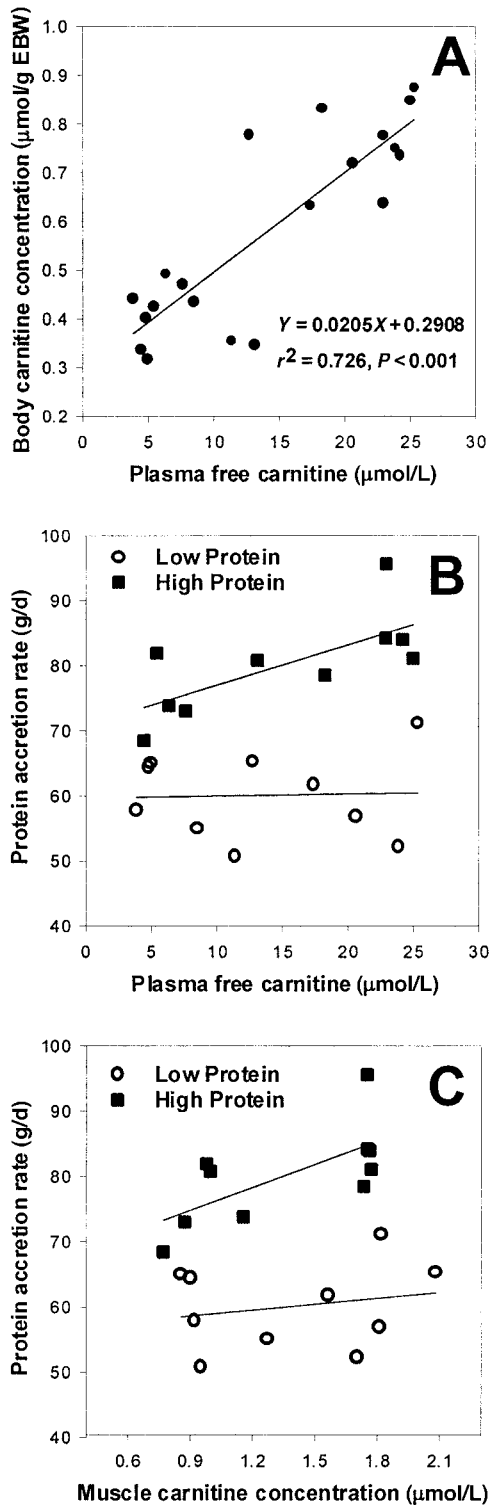


FIGURE 3 Linear relationships between plasma free carnitine and body carnitine [per gram empty body weight (EBW)] (A), plasma free carnitine and protein accretion (B; high protein level, $Y = 0.618X + 70.85$, $r^2 = 0.500$, $P < 0.023$; low protein level, $Y = 0.028X + 59.74$, $r^2 = 0.001$, $P = 0.93$), and muscle carnitine and protein accretion (C; high protein level, $Y = 11.69X + 64.3$, $r^2 = 0.457$, $P < 0.032$; low protein level, $Y = 3.02X + 55.9$, $r^2 = 0.045$, $P = 0.55$) of 20-kg pigs. Plasma free carnitine was measured 2 h after previous meal (on d 10).

dietary fat, and thus nitrogen utilization was affected. The improved biological value of nitrogen in the carnitine-supplemented group suggests that more dietary amino acids were used

for body protein synthesis rather than for energy. Collectively, these observations show that carnitine improved nitrogen utilization under ME-limited conditions.

Nutrient accretion and body composition. Owen et al. (1996) found that carcass lipid and daily lipid accretion were reduced, but carcass protein and daily protein accretion were unaffected by L-carnitine supplementation in weaning pigs. A similar trend was found in older pigs (Owen et al. 1993 and 1994) in that L-carnitine improved carcass characteristics (i.e., reduced lipid accretion rate and backfat thickness) and feed efficiency, but did not affect ADG in growing-finishing pigs. By contrast, lipid accretion was not reduced, but daily protein accretion was increased in our study. This discrepancy is possibly due to differences in energy status and lysine to ME ratio between the two experiments. Owen et al. (1996) offered diets containing 5% soy oil ad libitum to weaning pigs from 6 to 20 kg. Even if the L-carnitine-supplemented group might have increased β -oxidation, decreased lysine degradation and improved ME for protein synthesis, the unsupplemented group could have increased lysine and ME available for protein synthesis by increasing feed intake (i.e., ad libitum consumption). Therefore, it is possible that L-carnitine did not affect daily protein accretion and ADG, but decreased average daily feed intake in their research. In contrast, we offered 85% of estimated ad libitum energy intake (0.63 or 0.84 g lysine/MJ ME for the low protein or high protein level, respectively) to growing pigs from 18 to 22 kg. Lower energy status (by restriction) was implemented to amplify putative L-carnitine effects on protein accretion. With this constraint, two possible mechanisms are plausible. One is that L-carnitine increased β -oxidation of fatty acids and supplied more ME for protein synthesis. The other is that lysine degradation was reduced by the increased ME from fat such that more dietary amino acids could be directed toward protein synthesis. The latter is supported by the increase in the biological value of N observed when L-carnitine was supplemented in the present study. Increased daily protein accretion and reduced body fat composition due to L-carnitine supplementation support both possibilities. Bohles et al. (1984b) reported similar findings in that L-carnitine supplementation of mini-pigs during total parenteral nutrition with lipid emulsion increased lipolysis, oxidation of fatty acids, energy gain from infused fat and N balance. In particular, the branched-chain amino acids in the muscle of the miniature piglets were increased with L-carnitine (Bohles et al. 1984a). In a similar manner, Ji et al. (1996) concluded that L-carnitine altered intermediary metabolism and reduced body fat without changing growth rate in Atlantic salmon.

Even though pigs fed both of our basal diets were marginally restricted in daily ME, the high protein diet was relatively more restricted by a low ME/lysine ratio. Indeed, this was evidenced by the higher urinary N excretion from amino acid degradation in pigs fed the high protein diet. For this reason, it was expected that the addition of L-carnitine to the high protein diet would preferentially improve growth performance and nutrient accretion rate compared with that in low protein level. Even though the responses to carnitine in the high protein diet were larger than those from the low protein diet, no significant interaction (protein level \times L-carnitine) was detected for any criteria by ANOVA. This implies that pigs fed the low protein diet also were restricted sufficiently in daily ME for L-carnitine to affect all criteria in this study. Regression analysis (Fig. 3B and C), however, did support deviate responses for the two protein levels, with greater apparent responses to carnitine in the high protein diet compared with the low protein diet. Taken together, our data support the hypothesis that supplemented L-carnitine improves the effi-

ciency of nitrogen utilization by 20-kg pigs fed fat-containing diets that are limited in ME. Thus, L-carnitine increased rate of gain in general and protein accretion rate in particular.

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