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ORIGINAL ARTICLE

Effects of feeding L-carnitine to gilts through day 70 of gestation on litter traits and the expression of insulin-like growth factor system components and L-carnitine concentration in foetal tissues

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Summary

We investigated the influence of supplemental L-carnitine on foetal blood metabolites, litter characteristics, L-carnitine concentration in skeletal muscle and insulin-like growth factor (IGF) axis components in foetal hepatic and skeletal muscle tissues at day 40, 55 and 70 of gestating gilts. A total of 59 gilts (body weight = 137.7 kg) received a constant feed allowance of 1.75 kg/day and a top-dress containing either 0 or 50 ppm of L-carnitine starting on the first day of breeding through the allotted gestation length. Foetuses from the gilts fed diets with L-carnitine tended to be heavier ($p = 0.06$) and the circulating IGF-II tended to be lower ($p = 0.09$) at day 70, compared with the foetuses from the control gilts. Insulin-like growth factor-I messenger RNA (mRNA) was lower ($p = 0.05$) in hepatic tissue in the foetuses collected from gilts fed L-carnitine. Free and total carnitine concentration increased ($p < 0.05$) in the skeletal muscle from the foetuses collected from gilts fed supplemental L-carnitine. This study showed that L-carnitine had beneficial effects on the average foetal weight at day 70 of gestation, associated with changes in the foetal IGF system.

Introduction

Feeding multi-parous sows supplemental L-carnitine during gestation has been shown to increase their body weight (BW) gain (Musser et al., 1999; Ramanau et al., 2002), last rib backfat (Musser et al., 1999), plasma insulin-like growth factor-I (IGF-I) (Musser et al., 1999; Doberenz et al., 2006) and plasma IGF-II (Doberenz et al., 2006). In addition, supplementing L-carnitine to gestating sows may increase total number of pigs born and born alive (Ramanau et al., 2004; Birkenfeld et al., 2005), decrease the number of stillborn pigs (Musser et al., 1999; Doberenz et al., 2006), and increase the average pig (Eder et al., 2001; Ramanau et al., 2002, 2004) and litter weight at birth (Ramanau et al., 2002, 2004; Doberenz et al., 2006).

We have previously reported no change in BW, backfat, estimated protein mass, estimated fat mass or circulating IGF-I in gilts fed L-carnitine. But, we did observe changes in the IGF system components in the endometrium of gilts fed supplemental

L-carnitine (Brown et al., 2007). Therefore, the objective of this study was to determine the effects of supplemental L-carnitine on litter characteristics, concentration of carnitine components in foetal muscle and the expression of the IGF axis components in foetal tissues.

Materials and methods

Animals and treatments

All animal procedures used in this study were reviewed and approved by the Kansas State University Animal Care and Use Committee. Fifty-nine gilts (PIC: Franklin, KY; L327 × 1050; BW = 137.7 kg; 190 days of age) were artificially inseminated (PIC; MQ 280) 12, 24 and 36 h after the onset of their second oestrus. Day one was considered 12 h after the first insemination. Gilts were housed in individual crates (1.83 × 0.55 m) in an environmentally-controlled gestation barn at the Kansas State University Swine Teaching and Research Center from breeding until either day 39.5, 54.5 or 69.5 of gestation. Gilts were allowed *ad libitum* access to water and randomly allotted one of the two dietary treatments and one of three harvesting dates; based upon weight at breeding. All gilts were fed a corn–soybean meal-based gestation diet (Table 1) once daily (1.75 kg/day; as-fed basis) and received a 50-g ground corncob top-dress containing either 0 mg of Carniking 10 per 1.75 kg of diet (control group, $n = 30$) or 88 mg of Carniking 10 (10% L-carnitine, Lonza Group, Allendale, NJ, USA) per 1.75 kg of diet (L-carnitine group, $n = 29$) from day 1 to 39, 54, or 69 of gestation. The gestation diet was formulated to meet or exceed National Research Council (NRC) (1998) nutrient requirement estimated based on NRC models to be slightly above the requirements for maintenance and foetal growth. Foetal and uterine gain throughout pregnancy was predicted to be 25 kg with gilt maternal gain predicted at an additional 13.6 kg (Aherne and Kirkwood, 1985; Williams et al., 1985).

Harvesting protocol and collection of samples

Harvest was completed on gilts on either day 40, 55 or 70 of gestation. Fifteen hours before harvest, gilts were transported from the Kansas State University Swine Research and Teaching Center to the Kansas State University Meat Laboratory where sample collections were performed 24 h after the last feeding. Gilts were allowed *ab libitum* access to water until harvest. Gilts were harvested by electrical stunning followed by ex-sanguination. A mid-lateral incision was made to gain access to the abdominal cavity. The ovarian pedicles and uterine stump, at the level of the cranial cervix were cut and the uterus removed. Once the uterus was removed, the number of foetuses was determined in both horns. Foetal pigs were removed under aseptic conditions and rapidly transported to our laboratory for processing.

Foetal blood collection, weights and lengths

Individual foetuses were weighed and total litter weight was calculated as the sum of the individual foetus weights per litter. The crown to rump length was measured and foetal blood was collected from the heart of each foetus and pooled with the other foetuses in the litter for determination of foetal plasma IGF-II [IGF-II RIA IGF-binding proteins (IGFBP) Blocked, 022-IGF-R30; American Laboratory Products, Windham, NH, USA].

Table 1 Diet composition fed during gestation (as-fed basis)*

Item	Amount
Ingredient, %	
Corn	81.22
Soybean meal, 46.5% CP	14.55
Monocalcium phosphate, 21% P	2.03
Limestone	1.05
Salt	0.50
Vitamin premix†	0.25
Trace mineral premix†	0.15
Sow add pack†	0.25
	100.00
Calculated analysis	
Lysine, %	0.65
ME, Mcal/kg	3.27
CP, %	13.7
Ca, %	0.85
P, %	0.75
Available P, %	0.48

CP, Crude Protein; ME, metabolizable energy.

*Gestation feeding levels of 1.75 kg per day, with a 50-g ground corncob top-dress containing 0 mg of Carniking 10 per 1.75 kg of diet (control, $n = 30$) or 88 mg of Carniking 10 per 1.75 kg of diet (L-carnitine, $n = 29$) [Carniking 10; 10% L-carnitine, Lonza Group, Allendale, NJ] from 1 to day 39, 54 or 69 of gestation. Predicted L-carnitine content of basal diet was 43.75 ppm (Jacobs, 2002).

†Supplied per kilogram of diet: 11 025 IU of vitamin A, 1,654 IU of vitamin D_3, 55.1 mg of niacin, 44.1 IU of Vitamin E, 33.1 mg of pantothenic acid, 9.9 mg of riboflavin, 4.4 mg of vitamin K (menadione) and 0.04 mg of vitamin B_{12}. 165 mg of Zn (oxide), 165 mg of Fe (sulfate), 39.7 mg of Mn (oxide), 16.5 mg of Cu (sulfate), 0.30 mg of I (as Ca iodate), and 0.30 mg of Se (as Na selenite), 551.3 mg of choline, 15.2 mg pyridoxine, 1.65 mg of folic acid and 0.22 mg of biotin.

Sample preparation and RNA isolation

Semi-tendinosus muscle from the left hind limb and hepatic tissue from the left lobe was excised from each foetus, individual identity preserved and immediately snap-frozen in liquid nitrogen and stored at −80 °C in an Ultra Low Freezer. Total RNA was isolated from foetal hepatic and skeletal muscle tissue by using the RNeasy Mini Kit (Qiagen, Valencia, CA, USA). The concentration of RNA was determined by absorbance at 260 nm. Electrophoresis of total RNA through a 1% agarose-formaldehyde gel followed by ethidium bromide staining to allow visualization of 28S and 18S ribosomal RNA (rRNA) was carried out to assess the integrity of RNA. One microgram of total RNA was then reverse-transcribed to produce the first-strand complementary DNA (cDNA) using TaqMan reverse transcriptase (Applied Biosystems, Foster City, CA, USA) following the protocol recommended by the manufacturer. Random hexamers were used as primers in cDNA synthesis.

Real-time quantitative PCR

Real-time quantitative-PCR was used to measure the quantity of mRNA for IGF-I, IGF-II, IGFBP-3, IGFBP-5 and 18S rRNA in total RNA isolated from hepatic and skeletal muscle tissues. Measurement of the relative quantity of cDNA was carried out using TaqMan Universal PCR Master Mix (Applied Biosystems), 900 nM of the appropriate forward and reverse primers, 200 nM of the appropriate TaqMan detection probe, and 1 μl (0.5 μg cDNA) of the cDNA mixture. Sequences for primers and probes for IGF-I, IGF-II, IGFBP-3 and IGFBP-5 are presented in Table 2. Commercially available eukaryotic 18S rRNA primers and probes were used as an endogenous control (Applied Biosystems; GenBank Accession no X03205). Assays were performed in an ABI Prism 7000 sequence detection system (Applied Biosystems) using thermal cycling parameters recommended by the manufacturer (50 cycles of 15 s at 95 °C and 1 min at 60 °C). Relative expressions of mRNA for IGF-I, IGF-II, IGFBP-3 and IGFBP-5 were normalized to the 18S rRNA endogenous control and expressed in arbitrary units.

Carnitine and acyl-carnitine analyses

Tissue samples were homogenized in perchloric acid and free carnitine and acyl-carnitines were extracted with perchloric acid as described by Lin and Odle (2003). The extracted carnitine and acyl-carnitines were determined using the enzymatic radioisotope method of McGarry and Foster (1976) with minor modifications as described by Bhuiyan et al. (1992).

Table 2 Primers and probes used for real-time quantitative PCR

Gene	GenBank accession number	Sequence
IGF-I		
Forward	M31175	TCTTCTACTTGGCCCTGTGCTT
Reverse		GCCCCACAGAGGGTCTCA
Probe		6FAM-CCTTCACCAGCTCTGCCACGGC-TAMRA
IGF-II		
Forward	X56094	CCGGACAACTTCCCCAGATA
Reverse		CGTTGGGCGGACTGCTT
Probe		6FAM-CCCGTGGGCAAGTTCTTCCGC-TAMRA
IGFBP-3		
Forward	AF085482	AGCACGGACACCCAGAACTT
Reverse		CGGCAAGGCCCGTATTC
Probe		6FAM-TCCTCTGAGTCCAAGCGCGAGA-TAMRA
IGFBP-5		
Forward	U41340	GGCAGAGGCCGTGAAGAAG
Reverse		CAGCTCCCCCCACGAACT
Probe		6FAM-CCGCAGAAAGAAGCTGACCCAGTCC-TAMRA

IGF, insulin-like growth factors; IGFBP, insulin-like growth factors binding proteins.

Statistical analyses

Foetal weights, crown rump length and IGF-II blood concentration data were analysed as a 2×3 factorial arrangement with the MIXED Procedure of SAS (SAS Institute Inc., Cary, NC, USA). Fixed model effects included treatment, day of harvest, and their interaction. Random effects included gilt (treatment). Carnitine tissue concentrations were analysed as a 2×2 factorial arrangement with the MIXED Procedure of SAS, as no foetal tissue was available on day 40. Fixed model effects included treatment, day of harvest and their interaction. Random effects included gilt (treatment) and Kenward–Roger adjustment was used for the degrees of freedom .

The Fisher's Exact method was used to determine p-values of a chi-square statistic between differences in the number of litters that were detectable for IGF-II for L-carnitine and control-fed gilts using the IGF-II RIA (Higgins, 2004).

For all genes evaluated, mRNA concentrations were analysed as a 2×3 factorial arrangement with the MIXED Procedure of SAS. Fixed model effects included treatment, day of harvest and their interaction. Kenward–Roger adjustment was used for the degrees of freedom. The significance was declared at $p < 0.05$ unless noted otherwise.

Results

Litter characteristics

Total foetus number and total litter weight were not different ($p > 0.05$) at day 40, 55, or 70 of gestation for the gilts fed L-carnitine or the control diet (Table 3). In addition, no differences ($p > 0.05$) were observed in foetus number or crown rump length between the right uterine or left uterine horn. As gestation length increased, total litter weight, average foetal weight, crown rump length and IGF-II increased ($p < 0.05$), but total foetus number and foetus number in the right and left uterine horn decreased ($p < 0.05$). At day 70 of gestation, foetuses from the gilts fed L-carnitine tended to be heavier (+8%, $p = 0.06$) than the foetuses from the control fed gilts. The number of litters where IGF-II was detectable was greater in the foetuses from the gilts fed no supplemental L-carnitine at day 55 (+55%, $p = 0.06$) and day 70 (+40%, $p = 0.09$). Foetuses from gilts fed L-carnitine had numerically lower (−30%, $p = 0.09$) plasma IGF-II concentrations, compared with the foetuses from the gilts fed the control diet at day 70 of gestation. No differences were observed at day 55 of gestation ($p = 0.61$) in IGF-II concentration between the two treatments.

RNA analysis

Overall, IGF-I mRNA was lower (−83%, $p = 0.05$) in the hepatic tissue of the foetuses, collected from gilts fed supplemental L-carnitine (Table 4). At day 40 of gestation, hepatic tissue IGF-I was only 35% of the control level ($p = 0.01$) in foetuses collected from gilts fed L-carnitine compared to the foetuses collected from the gilts fed the control diet. Foetal hepatic tissue IGF-II, IGFBP-3 and IGFBP-5 mRNA expression was not influenced ($p > 0.05$) by dietary treatment. As day of gestation increased, foetal hepatic IGFBP-3 mRNA levels increased ($p < 0.01$). No differences ($p > 0.05$) in IGF-I, IGF-II, IGFBP-3 and IGFBP-5 mRNA expression were observed in foetal muscle between dietary treatments. A main effect of day was observed for foetal muscle IGF-I ($p = 0.02$) and IGFBP-3 ($p = 0.03$) mRNA. As days of gestation increased from 40 to 70, foetal muscle IGF-I mRNA expression increased. As the days of gestation increased from 40 to 55, IGFBP-3 mRNA decreased and from 55 to 70 day, IGFBP-3 mRNA

Table 3 Effects of L-carnitine on foetal weight, number, length and circulating IGF-II*

	Day of Gestation									
	40		55		70					
	L-carnitine, ppm†							p-value		
Item	0	50	0	50	0	50	SED	Trt‡	Day§	Trt × Day
No. gilts	9	10	10	10	10	10				
Litter										
Total litter weight, g	136.7	122.4	901.0	883.9	2484.5	2657.1	188.20–183.18	0.66	<0.01	0.71
Average foetal weight, g¶	9.5	8.4	74.6	69.2	217.7	236.6	9.95–10.30	0.48	<0.01	0.19
Total foetus no.	14.0	14.1	12.1	12.8	11.5	11.4	1.20–1.23	0.74	0.01	0.89
Foetus no. right horn	7.4	6.9	5.7	6.1	5.8	6.2	0.75–0.77	0.83	0.05	0.63
Foetus no. left horn	6.6	7.1	6.3	6.7	5.7	5.1	0.74–0.76	0.80	0.02	0.54
Crown rump length, cm	5.3	5.2	12.0	11.6	16.8	16.8	0.32–0.33	0.44	<0.01	0.80
Detectable IGF-II**	‡‡	‡‡	9	4	10	6				
IGF-II, ng/ml††	‡‡	‡‡	16.3	14.5	22.9	17.6	3.04–3.54	0.14	0.05	0.45

IGF, insulin-like growth factors; IGFBP, insulin-like growth factors binding proteins.

*Data were analysed as a 2 × 3 factorial arrangement and are least squares means and SED.

†Gestation feeding levels of 1.75 kg per day, with a top-dress providing 0 mg of Carniking 10 per 1.75 kg of diet or 88 mg of Carniking 10 per 1.75 kg of diet.

‡Treatment (Trt).

§Day of gestation (day).

¶Day 70 $p = 0.06$.

**The number of litters where IGF-II was detectable. Data analysed using a chi-squared statistic (day 55, $p = 0.06$ and day 70, $p = 0.09$).

††Day 70, $p = 0.09$.

‡‡No data available.

levels increased similar to the levels at day 40 of gestation.

Carnitine and acyl-carnitine

Free and total carnitine concentrations were higher ($p < 0.05$ and $p < 0.02$, respectively; Table 5) in the skeletal muscle tissue of foetuses collected from gilts fed supplemental L-carnitine. No differences ($p > 0.05$) were observed for short-chain or long-chain acyl-carnitine concentrations. As day of gestation increased from 55 to 70, there was an increase ($p < 0.01$) in free, long-chain acyl and total carnitine concentrations in the skeletal muscle.

Discussion

Recent studies have reported beneficial effects of feeding supplemental L-carnitine to sows. Waylan et al. (2005) found that at day 55 of gestation, the number of foetuses was greater in the sows supplemented with L-carnitine in gestation compared with the sows fed the control diet. Ramanau et al. (2004) and Birkenfeld et al. (2005) found similar results when sows were fed supplemental L-carnitine during gestation. These researchers found an increase in the total number of pigs born and total number of pigs born alive (Ramanau et al., 2004; Birkenfeld et al., 2005). In addition, Musser et al. (1999) and Doberenz et al. (2006) found a reduction in the number of stillborn pigs at birth. This data suggests that L-carnitine may aid in improving conception rate or reducing embryo mortality. In contrast to these findings, the present study shows no differences in the total number of foetuses at day 40, 55, or 70 of gestation. The studies that observed an increase in the total number of pigs also observed the increase in both gilts and sows. In our study, we observed no differences in the total number of foetuses in gilts. The exact mechanism behind the observed increase in total number of pigs born in the other studies is unclear. L-carnitine appears to affect gilts and sows differently (Eder et al., 2001; Ramanau et al., 2005) and one could speculate that in older animals L-carnitine may be allowing greater nutrient uptake in a mature uterus which allows for enhanced foetal growth and development and increased embryo survival. But, in the study made by Doberenz et al.

Table 4 Influence of L-carnitine supplementation to gestating gilts on foetal hepatic and skeletal muscle tissue mRNA relative abundance of IGF-system genes*

	Day of Gestation									
	40		55		70					
	L-carnitine, ppm¶							p-value		
Item	0	50	0	50	0	50	SED	Trt**	Day††	Trt × Day
No. gilts	9	10	10	10	10	10				
No. foetuses	9	10	10	10	10	10				
Hepatic†										
IGF-I‡	1.74	0.61	0.62	0.58	0.58	0.67	0.437–0.449	0.05	0.24	0.17
IGF-II	14 504	11 748	13 930	13 776	10 408	10 316	3,055–3,139	0.67	0.51	0.82
IGFBP-3	27.90	37.76	61.33	63.46	60.66	68.01	11.51–11.83	0.34	0.0004	0.89
IGFBP-5	2.83	2.63	3.58	3.28	3.93	4.01	0.929–0.955	0.80	0.19	0.96
Skeletal Muscle§										
IGF-I	12.75	10.82	23.76	9.98	54.64	56.76	12.71–24.36	0.74	0.02	0.89
IGF-II	6814	8312	14 456	3876	40 326	76 556	27 467–28 220	0.67	0.32	0.62
IGFBP-3	149.16	184.34	70.98	31.86	131.78	235.12	77.83–79.96	0.47	0.04	0.44
IGFBP-5	393.36	312.20	257.80	209.40	213.53	275.66	105.37–108.26	0.72	0.23	0.61

IGF, insulin-like growth factor; IGFBP, insulin-like growth factor binding protein.

*Data were analysed as a 2 × 3 factorial arrangement. Data are least squares means and SED and presented as arbitrary units expressed in millions.

†Total RNA was isolated from the left lobe of the liver at the median conceptus from each uterine horn.

‡Day 40, $p = 0.01$.

§Total RNA was isolated from semi-tendinosus muscle from the left hind limb at the median conceptus from each uterine horn.

¶Gestation feeding levels of 1.75 kg per day, with a top-dress containing 0 mg of Carniking 10 or 88 mg of Carniking 10 per 1.75 kg of diet.

**Treatment (Trt).

††Day of gestation (Day).

Table 5 Influence of L-carnitine supplementation to the dam on foetal skeletal muscle tissue L-carnitine concentration*

	Day of Gestation							
	55		70					
	L-carnitine, ppm†					p-value		
Item	0	50	0	50	SED	Trt‡	Day§	Trt × Day
No. samples	6	6	6	6				
Muscle tissue levels, nmol/g								
Free carnitine	29.1	32.8	73.9	98.3	9.74	0.05	<0.01	0.15
Short-chain acyl-carnitine	5.8	6.5	3.1	6.2	2.71	0.33	0.46	0.54
Long-chain acyl-carnitine	2.9	3.2	6.0	7.7	1.47	0.34	<0.01	0.51
Total carnitine	37.7	42.5	83.0	112.4	9.83	0.02	<0.01	0.09

SED, standard error of the difference of the means.

*Data were analysed as a 2 × 2 factorial arrangement and values are least squares means and SED.

†Gestation feeding levels of 1.75 kg per day, with a top-dress providing 0 or 88 mg of Carniking 10 per 1.75 kg of diet.

‡Treatment (Trt).

§Day of gestation (Day).

(2006), L-carnitine was given 21 days before insemination and throughout pregnancy to sows of third parity and litter number was not increased. One could speculate that if L-carnitine was fed through successive gestation periods, this may affect follicular development. Unlike our study, the study conducted by Birkenfeld et al. (2005) fed L-carnitine through successive gestation periods may have allowed L-carnitine to play a role in follicular development. In addition, the number of gilts involved in this study may not have been sufficient to determine differences in the number of foetuses.

Recent studies have reported increased pig and litter birth weights when sows are supplemented with L-carnitine (Ramanau et al., 2002, 2004). At day 70 of gestation, we observed a trend for increased average foetal weight in foetuses collected from gilts fed diets containing supplemental L-carnitine, compared with the foetuses collected from the gilts fed the control diet. In addition, other researchers have found increased muscle fiber numbers in offspring from sows fed diets with supplemental L-carnitine (Musser et al., 1999). Researchers have hypothesized that the heavier piglets born from sows fed supplemental L-carnitine is because of an increased intrauterine nutrient supply of glucose (Ramanau et al., 2002), an increase in glucose transporter-1 (Doberenz et al., 2006), or an increased maternal IGF-I (Musser et al., 1999) via action on the placenta thereby possibly improving placental function and subsequently nutritional supply to the foetus (Doberenz et al., 2006). Therefore, the increased nutrient supply and glucose, in turn, may increase IGF-I in the foetus. The role of IGF-I in normal growth and development has been well documented and plays an important role in muscle cell proliferation (Florini et al., 1991) and increased birth weight (Hills et al., 1996). Thus, the elevated levels of maternal IGF-I found by Musser et al., (1999) may have improved the nutrient supply to the foetus, increasing IGF-I in the foetus, and improving muscling in these pigs. But, our data (Brown et al., 2007) and that of Waylan et al. (2005) suggests that the average foetal weights were heavier from gilts fed L-carnitine was not because of increased maternal IGF-I. Therefore, an understanding of the changes in the IGF system in foetal tissues will aid in our comprehension of foetal muscle development and the trend for improved foetal weight we observed at day 70 of gestation because of supplemental L-carnitine.

Insulin-like growth factor-I and -II are the key regulators involved in foetal muscle development. In muscle cells, IGF-I promotes muscle proliferation (Florini et al., 1991), while IGF-II can promote muscle differentiation (Moses et al., 1980). Waylan et al. (2005) found when mono-nucleated porcine embryonic myoblasts collected from foetuses of dams fed supplemental L-carnitine differentiate into primary muscle fibers, IGF-II mRNA expression decreases. This would suggest that supplementation of L-carnitine to the dams allow a greater myoblast proliferation in foetuses because of lower levels of IGF-II mRNA expression. We can hypothesize that this may allow a greater number of porcine embryonic myoblasts to differentiate into additional muscle fibers or may allow more DNA fusion during myoblast differentiation, therefore, increasing muscle size. The average foetus weight from the gilts fed

L-carnitine was 18.9 g heavier than the average foetus weight from the gilts fed the control diet at day 70 of gestation. In our study, we found a tendency toward decreased levels of circulating IGF-II in foetuses collected from gilts fed supplemental L-carnitine and undetectable levels of IGF-II, in half of those foetuses. These data suggest IGF-II may play a role in the increased weight we observed at day 70 of gestation in foetuses collected from gilts fed supplemental L-carnitine. Feeding L-carnitine decreased circulating foetal IGF-II and we hypothesize that it may have increased foetal IGF-I, but this was not measured. Therefore, feeding L-carnitine may have increased cell proliferation producing heavier foetuses and caused a delay in muscle cell differentiation late in gestation.

It has also been noted that levels of amino acids were greater in pigs fed supplemental L-carnitine suggesting either increased protein accretion or reduced protein break down (Bohles and Lehnert, 1984; Alvestrand et al., 1990; Owen et al., 2001). We found supplementing the gilt L-carnitine increased the carnitine concentration in the foetal skeletal muscle which may have increased the supply of amino acids allowing for greater protein accretion resulting in heavier foetuses at day 70 of gestation. In addition, these data offers compelling evidence that dietary L-carnitine fed to gestating gilts can be absorbed and ultimately transported to the developing foetus to be stored in tissues such as skeletal muscle. This finding agrees with a recent study of Birkenfeld et al. (2006), which found increased plasma and carcass concentrations in newborn piglets of sows, supplemented with L-carnitine.

In our study, we observed a decrease in hepatic IGF-I mRNA expression in foetuses collected from gilts fed supplemental L-carnitine. This is in contrast to results reported by Waylan et al. (2005) where the authors found no change in foetal hepatic IGF-I mRNA expression at day 55 of gestation. Previous studies using an ovine model show that maternal under-nutrition decreases hepatic IGF-I mRNA expression (Brameld et al., 2000). Our results are unexpected because for reasons of increased pig weight at birth thus; one can speculate supplementation of L-carnitine to the gilts and sows increases nutrient transfer to the foetus. Therefore, one would expect an increase in IGF-I mRNA expression because of increased nutrient levels. During foetal development, IGF-II is an important regulator because of its abundance in foetal tissues (Brown et al., 1986; Hill, 1990) and serum (Moses et al., 1980), but we observed no changes in IGF-II mRNA expression in foetal hepatic tissue. These unexpected findings observed in gene expression in hepatic tissue are not in agreement with our assumed role that L-carnitine plays in foetal development.

These results suggest L-carnitine supplementation to gestating gilts has beneficial effects on average foetal weight by day 70 of gestation, possibly observed because of its ability to reduce foetal IGF-II concentrations. Heavier foetal weights at day 70 of gestation from gilts fed L-carnitine was not because of maternal IGF-I (Brown et al., 2007). Therefore, the role of maternal IGF-I impact on foetal growth and development is unclear. The decreased expression of IGF-I in foetal hepatic tissues provides support for a change in IGF system components because of supplementation of L-carnitine to the gilt. Additional research in growth factor regulation is necessary to determine the effect of L-carnitine on biological mechanisms in the gestating and foetal pigs.

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