Prepartum fatty acid supplementation in sheep. IV. Effect of calcium salts with eicosapentaenoic acid and docosahexaenoic acid in the maternal and finishing diet on lamb liver and adipose tissue during the lamb finishing period[1]

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ABSTRACT: The objective of this study was to evaluate the effects of eicosapentaenoic acid (**EPA**) and docosahexaenoic acid (**DHA**) supplementation to ewes during late gestation on finishing lamb liver and adipose tissue fatty acid (**FA**) profile and gene expression. Lambs born from ewes supplemented with Ca salts of EPA + DHA, or palm FA distillate (**PFAD**) high in palmitic and oleic acid at 0.39% DM during the last 50 d of gestation were used. Lambs were weaned at 61 d of age and adapted to a high concentrate diet for 1.5 mo. After adaptation, 74 lambs (28 pens) were blocked by sex and BW and used in a 2 × 2 factorial arrangement of treatments using the factors of dam supplementation (**DS**) and lamb supplementation (**LS**) of Ca salts of EPA + DHA or PFAD at 1.48% DM. Lambs were slaughtered after 42 d and liver and adipose tissue collected for FA and gene expression analysis. Liver concentrations of EPA and DHA were greater ($P < 0.01$) with LS of EPA + DHA vs. PFAD during the finishing period. In adipose tissue, a lamb × dam interaction was observed for EPA ($P = 0.02$) and DHA ($P = 0.04$); LS of EPA + DHA increased EPA and DHA, but the increase was greatest in lambs born from ewes supplemented with PFAD. No lamb × dam treatment interactions were observed for gene expression in liver tissue ($P > 0.10$). Hepatic mRNA abundance of *hormone-sensitive lipase* (***HSL***; $P = 0.01$) was greater in lambs born from EPA + DHA ewes vs. lambs from PFAD ewes. mRNA expression of *stearoyl-CoA desaturase* ($P < 0.01$), *fatty acid synthase* ($P = 0.01$), *Δ^5-desaturase* ($P < 0.01$), and *Δ^6-desaturase* ($P < 0.01$) were decreased in liver of EPA + DHA lambs. A significant lamb × dam diet interaction was observed for elongation of *very long chain fatty acid 2* in adipose tissue ($P = 0.01$); lambs supplemented with the same FA as their dams had lower expression. Expression of *HSL* tended ($P = 0.08$) to be decreased in adipose of EPA + DHA lambs born from EPA + DHA ewes. The changes in mRNA expression suggest that lipogenesis decreased, and lipolysis increased in lamb liver with EPA + DHA vs. PFAD supplementation during the finishing period. In adipose tissue, changes suggest that lipogenesis decreased in lambs born from EPA + DHA supplemented dams and supplemented with EPA + DHA during the finishing period. In addition, these results suggest an interaction between supplementation of FA to dams during late gestation on lamb response of adipose tissue, but not liver, to FA supplementation during the finishing period.

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INTRODUCTION

Feeding fatty acids (**FA**) alters the FA profile of animal tissues (Shingfield et al., 2013). The omega-3 (n-3) FA eicosapentaenoic acid (**EPA**) and docosahexaenoic acid (**DHA**) function as bioactive molecules and can activate the transcription factor PPARα (Clarke, 2001). This activation increases the transcription of lipolytic genes and decreases the transcription of lipogenic genes, potentially increasing the utilization of FA for energy in liver tissue, which yields more energy than other metabolizable nutrients (Clarke, 2001). Although there is evidence for similar effects in ruminants (Bionaz et al., 2012), less is known about the effects of EPA and DHA on liver and subcutaneous adipose tissue gene expression in ruminants than in nonruminants. In addition, recent work has indicated that when supplemented in the diet as calcium (Ca) salts, EPA and DHA may have differential effects in subcutaneous adipose tissue and liver tissue (Coleman et al., 2018a).

Fetal programming is the process through which stimuli during the development of a fetus have lifelong effects on offspring (Godfrey and Barker, 2001). Fatty acids are one nutrient that may have fetal programming effects when supplemented during gestation because maternal supplementation alters the FA that are transferred to the fetus (Noble et al., 1978; Garcia et al., 2014a). In dairy cattle, supplementing multiparous cows with fats differing in FA profile [saturated fatty acids (**SFA**) vs. essential polyunsaturated fatty acids (**PUFA**)], but providing similar amounts of energy, increased birth weight compared with dams not supplemented with fat (Garcia et al., 2014a). The calves born from dams supplemented with SFA also had greater intakes and average daily gain through 60 d of age (Garcia et al., 2014b). In sheep, FA supplementation during gestation and lactation has been shown to affect offspring growth (Capper et al., 2006, 2007). When supplemented to dams during gestation, EPA and DHA alter offspring development through changes in metabolism in nonruminants (Mennitti et al., 2015), but little is known about their effects in ruminants. In beef cattle, supplementation of PUFA during the last third of gestation increased offspring BW compared with a diet with the same energy content but different FA profile (Marques et al., 2017). In sheep, supplementation of an enriched source of EPA and DHA during late gestation affected the FA profile of lamb plasma prior to weaning (Coleman et al., 2018a) and increased offspring performance during the finishing period (Carranza-Martin et al., 2018). However, the mechanisms behind these changes have not been elucidated. In addition, supplementing sources of n-3 FA to lambs during the finishing period alters the FA profile and gene expression of adipose and muscle tissue (Cooper et al., 2004; Urrutia et al., 2015, 2016). However, there are no studies that evaluate the interaction of FA supplementation to dams during late gestation and to offspring during the finishing period and the effects on offspring FA metabolism.

Therefore, we hypothesized that supplementation of Ca salts enriched with EPA and DHA to finishing lambs increases the concentrations of EPA and DHA in liver and subcutaneous adipose tissue, increases mRNA concentration of lipolytic genes, and decreases mRNA concentration of lipogenic genes in both tissues. In addition, we hypothesized that supplemental Ca salts enriched in EPA and DHA alter metabolism to a greater extent on lambs born from ewes supplemented with an enriched source of EPA and DHA during late gestation compared with lambs from ewes supplemented with a source of palmitic and oleic FA. The objective of this study was to evaluate the effect of supplementing Ca salts enriched with EPA and DHA on liver and adipose tissue FA profiles and gene expression in finishing lambs born from ewes supplemented with or without Ca salts enriched with EPA and DHA during the last 50 d of gestation.

MATERIALS AND METHODS

Experimental Design

This research study was conducted at the Sheep Center of the Ohio Agricultural Research

and Development Center, Wooster, OH (IACUC #2016A00000013). Seventy Hampshire × Dorset cross lambs (38 females and 32 males) were used in a 2 × 2 factorial arrangement of treatments. The lambs were born from ewes that were fed diets containing different Ca salts of FA during the last 50 d of gestation (first main factor: dam supplementation, **DS**). During the finishing period, lambs were fed diets with Ca salts of different FA (second main factor: lamb supplementation, **LS**).

Details of the experimental procedures of ewes and lambs pre-weaning have been described previously (Coleman et al., 2018a,b). Briefly, gestating ewes were blocked by BW and conception date into group pens with 3 animals per pen. The groups were randomly assigned to 1 of 2 treatments: 1) Ca salts of a palmitic FA distillate (**PFAD**) as a source of palmitic and oleic acids at 0.39% DM (EnerGII, Virtus Nutrition LLC, Corcoran, CA) and 2) Ca salts of EPA + DHA at 0.39% DM (StrataG113, Virtus Nutrition LLC, Corcoran, CA) (Table 1). After lambing, supplementation was ended, ewes and lambs were housed in individual lambing jugs for 12 to 24 h depending the time of the day they were born. After lambs had suckled from their dams, all ewes and lambs were placed onto pasture until weaning.

Details of the experimental animals and diet during the finishing period are fully described in the companion article (Carranza-Martin et al., 2018). Briefly, lambs were weaned at approximately 61 d of age and were then blocked by sex and size (large, medium, and small) based on initial finishing period BW and distributed into 28 pens (not evenly distributed with 2 or 3 lambs per pen). During the first 1.5 mo of the finishing period, the lambs were adapted to a high concentrate diet. After adaptation, lambs were fed diets that included PFAD or EPA + DHA at 1.48% DM. The amount of FA supplementation used was chosen to guarantee an intake of 18 mg per kg of metabolic BW ($BW^{0.75}$) of EPA and DHA. This dose was used based previous studies where metabolism was altered in humans (Bester et al., 2010). Calcium salts of PFAD were used as a control instead of a diet without fat to eliminate the confounding factor of diet energy density. Supplementation of the FA began after the adaptation period. Diets were formulated to meet or exceed NRC requirements for growing lambs (NRC, 2007) and contained 61.09% ground corn, 11.08% soybean meal, 24.08% soy hull, and 1.94% mineral and vitamin mix.

The diets were fed for 42 d, at which point lambs were slaughtered at the Ohio State University Department of Animal Sciences Meat Science Laboratory. Liver and subcutaneous adipose tissue samples were obtained from 1 lamb per pen postmortem (within 15 min) and snap-frozen using liquid N_2. Samples were stored at −80 °C until analysis. A liver sample was not obtained from 1 lamb from the small size block (DS:EPA + DHA; LS:PFAD).

Table 1. Fatty acid profile (% of total FA) of fat supplements fed to pregnant ewes during the last 50 d of gestation at 0.39% DM and to lambs during the finishing period at 1.48% DM

	Supplement[1,2]	
Fatty acid	PFAD	EPA + DHA
C12:0	0.62	0.12
C14:0	1.17	3.95
C14:1	0.02	0.12
C16:0	45.87	21.99
C16:1	0.20	7.40
C18:0	5.14	7.46
C18:1	36.27	17.44
C18:2	8.03	2.69
C20:0	0.37	0.34
C18:3	0.20	0.92
C20:5	0.13	9.18
C22:6	0	6.99
Other	1.98	21.88

[1]PFAD = EnerGII as a source of palmitic and oleic acids (Virtus Nutrition LLC, Corcoran, Ca); EPA + DHA = StrataG113 as a source of EPA and DHA (Virtus Nutrition LLC, Corcoran, Ca).

[2]Fatty acid profiles evaluated using the methods of Weiss and Wyatt (2003).

Tissue Analyses

The FA composition of liver and subcutaneous adipose tissue was determined using 300 mg (wet wt) of liver tissue and 150 mg (wet wt) of subcutaneous adipose tissue as per previous methods (O'Fallon et al., 2007) with slight modifications (Coleman et al., 2018a). Extraction of RNA from liver and subcutaneous adipose tissue was performed using a commercial lipid-specific extraction kit according to the manufacturer's protocol (74804 RNeasy Lipid Tissue Mini Kit, Qiagen, Hilden, Germany). The RNA from all samples was quantified using UV spectroscopy (Nanodrop Technologies) and qualitatively assessed using a BioAnalyzer 2100 and RNA NanoChip assay (Agilent Technologies). Two subcutaneous adipose tissue samples were not used for analysis due to RNA integrity numbers below 4.5. One sample was from DS:EPA + DHA and LS:PFAD, and the other one was from DS:PFAD and LS:EPA + DHA. Gene expression was determined using

a Nanostring nCounter XT Assay (Nanostring Technologies, Seattle, WA) and analyzed as described previously (Coleman et al., 2018a) for 27 genes (Table 2). These genes were chosen based on their involvement in FA uptake and release, FA synthesis and transcription factors that influence their expression, as well as genes for adipokines, hormone receptors, and inflammation (Lee et al., 2010; Bionaz et al., 2013; Contreras et al., 2017). Five housekeeping target genes were measured: *cyclophilin A*, *beta-actin*, *beta-2 microglobulin*, *glyceraldehyde 3-phosphate dehydrogenase and phosphoglycerate kinase 1*. There was an effect of treatment on the amount of mRNA for *cyclophilin A*, so it was removed and not included in the analysis and the data were only normalized to the geometric mean of the other 4 housekeeping target genes using the nSolver Analysis Software 3.0 (Nanostring Technologies, Seattle, WA).

Table 2. Gene names and GenBank accession number

Gene name[1]	Accession number
LPL	NM_001009394.1
ATGL	NM_001308576.1
HSL	NM_001128154.1
DGAT1	NM_001110164.1
DGAT2	XM_012096078.2
SCD	NM_001009254.1
Δ^5-desaturase	XM_012101996.2
Δ^6-desaturase	XM_015103138.1
ELOVL2	XM_012101293.2
ELOVL4	XM_015097304.1
ELOVL5	XM_012100862.2
FABP4	NM_001114667.1
FAS	XM_015098375.1
FATP1	XM_015095580.1
GIP receptor	XM_015100601.1
Ghrelin receptor	NM_001009760.1
Insulin receptor	XM_004008549.3
PPAR alpha	XM_012175774.2
PPAR beta/delta	XM_004018768.3
PPAR gamma	NM_001100921.1
RXR alpha	XM_012117960.2
Adiponectin	NM_001308565.1
Leptin	XM_004008038.3
Resistin	NM_001306111.1
COX-2	NM_001009432.1
5-LOX	XM_015104505.1
Beta-actin	NM_001009784.1
Beta-2 microglobulin	NM_001009284.2
Cyclophilin A	NM_001308578.1
GAPDH	NM_001190390.1
PGK1	NM_001142516.1

[1]LPL = lipoprotein lipase; ATGL = adipose triglyceride lipase; HSL = hormone-sensitive lipase; DGAT1 = diacylglycerol acyltransferase 1; DGAT2 = diacylglycerol acyltransferase 2; SCD = stearoyl-CoA desaturase; ELOVL2 = elongation of very long chain fatty acid 2; ELVL4 = elongation of very long chain fatty acid 4; ELOVL5 = elongation of very long chain fatty acid 5; FABP4 = fatty acid binding protein 4; FAS = fatty acid synthase; FATP1 = fatty acid transport protein 1; GIP = glucose-dependent insulinotropic polypeptide; PPAR = peroxisome proliferator activated receptors; RXR = retinoid × receptor; COX-2 = cyclooxygenase-2; 5-LOX = 5-lipoxygenase; GAPDH = glyceraldehyde 3-phosphate dehydrogenase; PGK1 = phosphoglycerate kinase.

Statistical Analyses

Data were analyzed as a randomized complete block design with a 2 × 2 factorial arrangement of treatments using the MIXED procedure (9.4, SAS Institute, Cary, NC) testing the fixed effects of dam treatment and lamb treatment and their interaction and the random effect of size (block). Pen was considered as the experimental unit. Sex and size (large, medium, and small) were included as blocks and removed when not significant ($P > 0.05$). Number of lambs at birth was included as a covariable and removed of the model because it was not significant. Statistical analysis of gene expression data was performed using the normalized data. Least square means and SE were determined using the LSMEANS statement in the MIXED procedure. Significance was set at $P \leq 0.05$, and tendencies were determined at $P > 0.05$ and $P \leq 0.10$.

RESULTS AND DISCUSSION

The basal diet fed to ewes during late gestation (DS) was formulated to meet the requirements of ewes during late gestation (NRC, 2007) and included the same amount of Ca salts. Lamb finishing diets (LS) were also formulated to meet or exceed nutrient requirements for growing lambs (NRC, 2007) and included Ca salts at the same amount. Therefore, the dam and lamb treatment diets only differed in the FA profile of the Ca salts. Thus, results of this experiment should be associated with the potential effects of supplemental EPA and DHA vs. PFAD, rather than the effects of fat itself. Although we recognize that the FA profile of the Ca salt were quite different, there are a limited number of commercial products available to provide FA to ruminants. The Ca salt of EPA and DHA utilized was the only product available to provide EPA and DHA at the time of the present study and was chosen with the Ca salt of PFAD to compare the effects of EPA and DHA vs. C16:0 and C18:1. In addition, similar models have been used previously in sheep (Carranza-Martin et al., 2018; Coleman et al., 2018a,b), beef cattle (Marques et al., 2017),

and dairy cattle (Garcia et al., 2014b, 2016). To our knowledge, this is the first study to investigate the effects of maternal supplementation with an enriched source of FA during late gestation on lamb liver and subcutaneous adipose tissue FA profiles and gene expression during the finishing period, and the interaction of supplementing an enriched source of FA to dams and then to their offspring during the finishing period.

Liver FA Concentration

A significant LS effect was observed for the concentration of C16:0 in liver tissue where concentrations were lower with EPA + DHA vs. PFAD supplementation during the finishing period ($P < 0.01$; Table 3). The greater concentration of C16:0 in liver tissue of lambs supplemented with PFAD during the finishing period is reflective of the greater dietary concentration of C16:0 in the Ca salt of PFAD compared with the Ca salt of EPA + DHA (Table 1). In addition, C16:0 is the primary product of FA synthesis by the enzyme FA synthase in mammals (Smith, 1994). Thus, the decrease in C16:0 with EPA + DHA supplementation during the finishing period may also indicate decreased FA synthesis in the liver.

Many of the C18:1 isomers were affected by LS during the finishing period. Concentrations of C18:1 *cis*-9 in liver tissue were greater with LS of PFAD ($P < 0.01$; Table 3). A significant LS effect was also observed for C18:1 *trans*-10 ($P < 0.01$) and C18:1 *trans*-11 ($P = 0.05$), where concentrations were greater in the liver tissue of lambs that were supplemented with EPA + DHA compared with PFAD. In addition, a significant LS effect was observed for C18:1 *trans 6,8* ($P < 0.01$), C18:1 *trans*-9 ($P < 0.01$), C18:1 *trans*-12 ($P = 0.05$), C18:1 *cis*-11 ($P < 0.01$), C18:1 *cis*-12 ($P = 0.04$), C18:1 *cis*-13 ($P < 0.01$), and C18:1 *cis*-15 ($P < 0.01$); concentrations of all of these FA were greater in liver tissue with LS of PFAD vs. EPA + DHA. Also, we observed a LS effect for the calculated C18:1 desaturase index, where the index was lower with LS of EPA + DHA ($P < 0.01$). The greater concentrations of C18:1 *cis*-9 in liver tissue of lambs supplemented with PFAD during finishing period might be due to the greater concentration of this FA in the Ca salt of PFAD vs. the Ca salt of EPA + DHA (Table 1). The FA C18:1 *cis*-9 can also be synthesized endogenously from C18:0 via desaturation by the enzyme stearoyl-CoA desaturase (**SCD**; Nakamura and Nara, 2004), and the C18:1 desaturase index can be used as an indicator of C18:1 *cis*-9 synthesis, where a lower index is associated with lower SCD activity and therefore lower synthesis of C18:1 *cis*-9 (Palmquist et al., 2004). Thus, the decrease in the concentration of C18:1 cis-9, as well as its desaturase index suggest that LS of EPA + DHA may decrease FA synthesis in the liver compared with PFAD supplementation. This potential decrease in synthesis of C18:1 *cis*-9 with EPA + DHA supplementation during the finishing period is associated with changes in liver *SCD* expression, as will be described later. In addition, both EPA and DHA alter biohydrogenation, resulting in the accumulation of biohydrogenation intermediates, primarily accumulation of *trans* isomers (Bauman and Griinari, 2003). Thus, changes in C18:1 *trans*-10, C18:1 *trans*-11 suggest that biohydrogenation was altered with supplementation of EPA + DHA vs. PFAD during the finishing period. Changes in other C18:1 isomers that were greater with PFAD supplementation during the finishing period may also be attributed to changes in biohydrogenation, as there is evidence that oleic acid may undergo *cis*/*trans* isomerization in the rumen, producing a variety of C18:1 intermediates (Kemp et al., 1984; Jenkins et al., 2006).

A DS effect was also observed for the C18:1 desaturase index; the index was lower in liver tissue of lambs born from dams supplemented with EPA + DHA during late gestation ($P = 0.05$). This suggests that DS with EPA + DHA during late gestation decreased the synthesis of C18:1 *cis*-9 in lamb liver tissue during the finishing period compared with maternal supplementation with PFAD. We hypothesized that maternal EPA + DHA supplementation would program offspring tissues toward decreased expression of lipogenic genes and that these changes would lead to differences in synthesis of FA. The lack of differences in other C18:1 isomers between DS treatments is not unexpected. Although maternal supplementation of FA does alter the transfer of FA to the fetus (Noble et al., 1978), supplementation of FA during late gestation alone would not directly alter rumen biohydrogenation pathways of the offspring.

The concentration of C18:2, linoleic acid, was significantly affected by LS ($P = 0.01$) and was greater in the liver tissue of PFAD lambs, whereas the concentration of C18:3, linolenic acid, was greater in liver of EPA + DHA lambs ($P < 0.01$). The concentration of C18:2 *cis*-12, *trans*-10 was greater ($P < 0.01$), whereas the concentration of C18:2 *cis*-9, *trans*-11 tended to be greater ($P = 0.08$) in the liver of PFAD lambs. The conjugated linoleic acid (**CLA**) desaturase index was also affected

Table 3. Liver fatty acid concentrations (% total fatty acid methyl esters[1]) of finishing lambs supplemented with Ca salts of EPA + DHA or palmitic fatty distillate acid (PFAD) at 1.48% and born from ewes supplemented with EPA + DHA or PFAD at 0.39% DM during the last 50 d of gestation[2]

Dam	PFAD		EPA + DHA			*P*-value[3]		
Lamb	PFAD	EPA + DHA	PFAD	EPA + DHA	SEM[4]	LS	DS	DS × LS
C12:0	0.04	0.06	0.02	0.05	0.02	0.16	0.37	0.67
C13:0	0.03	0.06	0.00	0.06	0.01	<0.01	0.11	0.22
C14:0 iso	0.02	0.03	0.00	0.05	0.02	0.05	0.99	0.11
C14:0	0.62	0.57	0.64	0.50	0.06	0.11	0.73	0.48
C14:1	0.09	0.05	0.08	0.03	0.02	0.01	0.37	0.47
C16:0 iso	0.13	0.15	0.16	0.17	0.02	0.20	0.08	0.61
C16:0	15.48	12.27	15.44	12.30	0.54	<0.01	0.99	0.94
C17:0	1.91	2.41	2.57	2.07	0.28	0.99	0.54	0.07
C17:1	0.79	0.27	0.96	0.24	0.10	<0.01	0.39	0.27
C18:0	21.86	20.12	22.09	21.95	0.84	0.21	0.17	0.28
C18:1 t6,8	0.44	0.24	0.44	0.22	0.06	<0.01	0.85	0.86
C18:1 t9	0.36	0.19	0.31	0.20	0.05	<0.01	0.65	0.49
C18:1 t10	3.75	6.37	2.98	5.56	0.84	<0.01	0.22	0.97
C18:1 t11	1.01	2.03	1.24	2.90	0.86	0.05	0.41	0.62
C18: t12	0.63	0.16	0.72	0.29	0.07	<0.01	0.09	0.80
C18:1 c9	17.94	9.00	16.60	4.76	1.46	<0.01	0.05	0.28
C18:1 c11	2.08	1.59	1.83	1.65	0.14	<0.01	0.40	0.14
C18:1 c12	0.18	0.07	0.23	0.14	0.05	0.04	0.18	0.86
C18:1 c13	0.55	0.18	0.50	0.20	0.04	<0.01	0.63	0.27
C18:1 c16	0.12	0.10	0.14	0.12	0.02	0.20	0.27	0.71
C18:1 c15	0.09	0.03	0.11	0.02	0.02	<0.01	0.70	0.19
C18:2	10.86	9.12	10.41	9.61	0.47	0.01	0.96	0.28
C20:0	0.23	0.23	0.29	0.24	0.03	0.34	0.17	0.31
C20:1	0.10	0.20	0.10	0.18	0.04	0.01	0.82	0.68
C18:3	0.26	0.36	0.25	0.37	0.04	<0.01	0.97	0.71
C18:2 c9,t11	0.24	0.19	0.22	0.17	0.03	0.08	0.57	0.91
CLA Other[5]	0.16	0.11	0.19	0.12	0.02	0.01	0.38	0.66
C18:2 c12,t10	0.21	0.12	0.23	0.15	0.03	<0.01	0.36	0.73
C21:0	0.24	0.15	0.22	0.15	0.03	<0.01	0.53	0.76
C22:0	0.19	0.71	0.38	0.93	0.18	<0.01	0.22	0.95
C20:3 n-6	1.40	0.60	1.21	0.46	0.20	<0.01	0.39	0.89
C20:3 n-3	0.15	0.12	0.16	0.10	0.01	<0.01	0.76	0.25
C22:1	0.08	0.15	0.09	0.17	0.01	<0.01	0.30	0.60
C20:4	11.42	5.28	11.49	5.93	0.40	<0.01	0.33	0.43
C20:5	0.57	4.83	0.52	5.29	0.23	<0.01	0.34	0.23
C24:0	0.16	0.16	0.17	0.22	0.02	0.30	0.14	0.20
C22:5	2.66	9.14	3.03	9.27	0.31	<0.01	0.38	0.67
C22:6	1.55	11.27	2.53	11.77	0.51	<0.01	0.12	0.61
Unidentified peaks	0.002	0.001	0.002	0.001	0.0001	<0.01	0.71	0.68
Total MUFA[5]	28.42	20.81	26.53	16.88	1.52	<0.01	0.05	0.47
Total PUFA[5]	29.48	41.13	30.26	43.26	1.30	<0.01	0.23	0.57
Total n-3[5]	5.17	25.70	6.51	26.83	0.89	<0.01	0.14	0.90
Total n-6[5]	24.32	15.44	23.73	16.41	0.71	<0.01	0.77	0.23
Total EPA and DHA	2.11	16.10	3.05	17.07	0.69	<0.01	0.14	0.98
Total saturated	42.10	38.06	43.21	39.86	0.93	<0.01	0.10	0.68
Total unsaturated	57.89	61.94	56.79	60.13	0.93	<0.01	0.10	0.68
14:1 desaturase index[6]	0.12	0.10	0.12	0.06	0.03	0.11	0.41	0.55
18:1 desaturase index[6]	0.45	0.30	0.43	0.17	0.04	<0.01	0.05	0.17
CLA desaturase index[6]	0.21	0.10	0.19	0.11	0.04	0.01	0.90	0.53
n-6/n-3	4.71	0.55	3.85	0.66	0.25	<0.01	0.05	0.01
CLA Total	0.61	0.42	0.64	0.44	0.08	0.004	0.68	0.95

[1]C15:0 iso and ante, C 15:0, C17:0 iso, C16:1 and C17:0 ante are not presented in the table because no differences due to DS, LS or DSxLS ($P \geq 0.14$), and we did not hypothesizes changes on those FA.

[2]PFAD = EnerGII as a source of palmitic and oleic acids (Virtus Nutrition LLC, Corcoran, Ca); EPA + DHA = StrataG113 as a source of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA; Virtus Nutrition LLC, Corcoran, Ca).

[3]*P*-values: DS = dam supplementation during the last 50 d of gestation; LS = lamb supplementation during the finishing period.

[4]SEM = most conservative SE for interaction was presented.

[5]CLA = conjugated linoleic acid; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; n-3 = omega-3; n-6 = omega-6.

[6]14:1 desaturase index = *cis*-9 C14:1/(C14:0 + *cis*-9 C14:1); 18:1 desaturase index = *cis*-9 C18:1/(C18:0 + *cis*-9 C18:1); CLA desaturase index = *cis*-9, *trans*-11 C18:2/(*cis*-9, *trans*-11 C18:2 + *trans*-11 C18:1.

by LS; the index was lowest in liver tissue of EPA + DHA lambs ($P = 0.01$). The changes in linoleic and linolenic acid are due to the differences in the concentrations of these FA between the 2 Ca salts (Table 1). In addition, biohydrogenation of linoleic acid results in the production of CLA isomers such as C18:2 *cis*-9, *trans*-11 and C18:1 *cis*-12, *trans*-10 (Jenkins et al., 2008). Therefore, increases in CLA isomers in liver tissue suggest that the increased linoleic acid in the Ca salt of PFAD may have modulated biohydrogenation pathways toward production of CLA intermediates. The CLA isomer C18:2 *cis*-9, *trans*-11 can also be synthesized in the body from C18:1 *tran*-11 via desaturation by SCD (Palmquist et al., 2004). As with the other desaturase indices described earlier, a lower value for the CLA desaturase index indicates a decreased production of C18:2 *cis*-9, *trans*-11 (Palmquist et al., 2004). This change is another indicator that hepatic activity of SCD may have been decreased with LS of EPA + DHA.

Only one DS × LS interaction was observed for FA in liver tissue; the ratio of n-6 to n-3 FA ($P = 0.01$; Table 3) was lowest in lambs that were born from dams supplemented with PFAD during late gestation and were supplemented with EPA + DHA during the finishing period. Lambs with DS and LS of EPA + DHA were the next lowest ratio, whereas DS of EPA + DHA and LS of PFAD was next. The greatest n-6:n-3 ratio was observed in lambs with DS and LS of PFAD. Hepatic concentrations of EPA (C20:5) and DHA (C22:6) were increased with LS of EPA + DHA vs. PFAD during the finishing period ($P < 0.01$). A significant LS effect was also observed for arachidonic acid (C20:4; **AA**) where concentrations in liver tissue were decreased with EPA + DHA vs. PFAD supplementation ($P < 0.01$). The changes in the these FA contributed to the significant increase and decrease in the total n-3 ($P < 0.01$) and omega-6 (n-6) FA ($P < 0.01$), respectively, in liver with LS of EPA + DHA. However, DS did not affect ($P \geq 0.12$) the concentrations of EPA, DHA, AA, total n-3, or total n-6 in lamb liver. Compared with a study by Demirel et al. (2004), the enrichment of EPA in liver tissue in the present study is lower, while enrichment of DHA in liver is similar. However, the differences in EPA and DHA enrichment between the present study and the aforementioned study by Demirel et al. (2004) are likely due to the fact that they supplemented fish oil with formaldehyde-treated whole linseed oil—the linseed oil would have provided a source of linolenic acid that could have been used to synthesize EPA and DHA, helping to increase their concentrations in tissues. However, the enrichment of EPA and DHA in liver tissue was greater in the present study compared with dairy cows that were fed fish oil or a saturated rumen-inert fat (Ballou et al., 2009). The greater concentrations observed with LS of EPA + DHA in our study is likely due to the use of a Ca salt of containing EPA and DHA, compared with the unprotected fish oil used by Ballou et al. (2009). To our knowledge, there are no other studies in ruminants investigating the effects of maternal supplementation with EPA + DHA on offspring liver tissue FA during the finishing period. However, our results suggest that maternal supplementation does not modulate the deposition of EPA and DHA in offspring liver tissue during the finishing period.

Subcutaneous Adipose Tissue FA Concentration

Lamb supplementation ($P = 0.05$) altered the concentrations of C10:0, C12:0, and C14:0 in subcutaneous adipose tissue; lambs supplemented with EPA + DHA had greater concentrations of these FA in their subcutaneous adipose tissue compared with lambs supplemented with PFAD (Table 4). The FA C10:0, C12:0, and C14:0 may be synthesized de novo in mammalian tissues (Schönfeld and Wojtczak, 2016). Thus, the increases in these FA might be may indicate increased FA synthesis in subcutaneous adipose tissue with LS of EPA + DHA. These results agree with previous data in our lab where the concentration of C10:0 was increased in subcutaneous adipose tissue of ewes after 1 mo of supplementation with EPA + DHA at 0.39% DM during late gestation (Coleman et al., 2018a). A study by Cooper et al. (2004) found that the percentages of C14:0 and C16:0 in subcutaneous adipose tissue were increased in lambs supplemented with fish oil or fish oil plus marine algae compared with supplementation of linseed oil, protected linseed oil (**PLS**), or PLS plus algae. In addition, Demirel et al. (2004) supplemented lambs with linseed, linseed plus fish oil, or Ca salts of palm oil and observed that lambs supplemented with linseed + fish oil had the highest concentrations of C16:0. However, supplementation of n-3 FA in young bulls using whole ground linseed decreased concentrations of C14:0, C15:0, and C16:0 in adipose tissue compared with bulls given no linseed, suggesting lower FA synthesis (Corazzin et al., 2013). In sheep, Urrutia et al. (2015) observed no difference in the concentrations of C12:0, C14:0, and C16:0 in subcutaneous adipose tissue with linseed or chia seed supplementation

Table 4. Subcutaneous adipose tissue fatty acid concentrations (% total fatty acid methyl esters[1]) of finishing lambs supplemented with Ca salts of EPA + DHA or palmitic fatty distillate acid (PFAD) at 1.48% and born from ewes supplemented with EPA + DHA or PFAD at 0.39% DM during the last 50 d of gestation[2]

Dam	PFAD		EPA + DHA			*P*-value[3]		
Lamb	PFAD	EPA + DHA	PFAD	EPA + DHA	SEM[4]	LS	DS	DS × LS
C10:0	0.13	0.14	0.14	0.16	0.01	0.05	0.10	0.64
C12:0	0.11	0.16	0.17	0.18	0.03	0.05	0.01	0.13
C13:0	0.02	0.02	0.02	0.03	0.003	0.06	0.01	0.20
C14:0	2.56	2.94	2.83	3.34	0.27	0.05	0.13	0.75
C15:0 iso	0.09	0.11	0.10	0.12	0.01	0.05	0.20	0.60
C14:1	0.04	0.07	0.10	0.10	0.02	0.49	0.01	0.51
C16:0	22.99	23.24	22.51	23.82	0.74	0.25	0.94	0.43
C18:0	19.73	14.81	15.28	13.81	1.96	0.04	0.07	0.24
C18:1 t6,8	0.02	0.005	0.005	0.001	0.013	0.34	0.35	0.55
C18:1 t9	—	0.04	0.06	—	0.04	0.84	0.84	0.15
C18:1 t10	8.66	9.92	7.65	8.68	1.19	0.29	0.29	0.92
C18:1 t11	0.31	0.65	0.53	0.56	0.19	0.80	0.68	0.36
C18:1 t12	—	0.03	—	—	0.01	0.19	0.19	0.19
C18:1 c9	27.84	28.81	33.03	30.25	1.99	0.47	0.01	0.14
C18:1 c11	1.04	1.5	1.09	1.11	0.06	0.06	0.50	0.12
C18:1 c12	0.22	0.14	0.18	0.16	0.02	0.05	0.68	0.14
C18:1 c13	0.08	0.08	0.07	0.09	0.02	0.60	0.93	0.63
C18:1 c16	0.04	0.05	0.04	0.04	0.02	0.57	0.70	0.76
C18:1 c15	0.08	0.07	0.08	0.10	0.02	0.81	0.60	0.33
C18:2	4.52	4.69	4.08	4.22	0.044	0.69	0.25	0.98
C20:0	0.08	0.07	0.05	0.05	0.01	0.60	0.02	0.53
C20:1	0.04	0.05	0.02	0.04	0.02	0.40	0.44	0.78
C18:3	0.34	0.46	0.46	0.48	0.05	0.23	0.25	0.47
C18:2 c9,t11	0.32	0.50	0.46	0.57	0.08	0.02	0.09	0.56
CLA other[5]	0.05	0.07	0.07	0.06	0.01	0.15	0.54	0.12
C18:2 c12,t10	0.07	0.06	0.06	0.05	0.01	0.19	0.09	0.73
C21:0	0.01	0.01	0.01	0.02	0.005	0.16	0.23	0.58
C22:0	0.01	0.02	0.003	0.02	0.005	<0.01	0.48	0.24
C20:3 n-6	0.02	0.05	0.04	0.03	0.02	0.44	0.88	0.20
C20:3 n-3	0.02	0.02	—	0.01	0.01	0.16	0.06	0.31
C22:1	0.02	0.01	0.04	0.04	0.02	0.75	0.33	0.82
C20:4	0.12	0.15	0.14	0.13	0.02	0.54	0.93	0.30
C20:5	0.01	0.17	0.02	0.11	0.02	<0.01	0.07	0.02
C24:0	0.003	0.02	0	0.01	0.01	0.14	0.37	0.55
C22:5	0.05	0.26	0.08	0.22	0.03	<0.01	0.85	0.14
C22:6	0.02	0.22	0.02	0.13	0.02	<0.01	0.07	0.04
Unidentified peaks	4.82	4.84	4.17	4.69	0.68	0.66	0.51	0.68
Total MUFA[5]	39.37	42.35	44.63	42.65	1.82	0.69	0.03	0.05
Total PUFA[5]	5.55	6.64	5.42	6.02	0.48	0.06	0.38	0.57
Total n-3[5]	0.47	1.13	0.58	0.95	0.08	<0.01	0.58	0.04
Total n-6[5]	5.07	5.51	4.84	5.06	0.44	0.40	0.39	0.79
Total EPA and DHA	0.03	0.39	0.04	0.25	0.04	<0.01	0.06	0.03
Total saturated	50.37	46.28	45.83	46.65	2.01	0.26	0.16	0.10
Total unsaturated	44.87	48.94	50.08	48.66	1.70	0.27	0.05	0.03
14:1 desaturase index[6]	0.02	0.02	0.03	0.03	0.01	0.90	0.01	0.23
18:1 desaturase index[6]	0.59	0.66	0.68	0.69	0.04	0.19	0.05	0.25
CLA desaturase index[6]	0.74	0.53	0.53	0.57	0.14	0.52	0.48	0.33
n-6/n-3	10.70	4.93	8.8	5.49	0.64	<0.01	0.11	0.01
CLA total	0.43	0.63	0.58	0.69	0.08	0.02	0.11	0.46

[1]C13:0 iso and ante, C14:0 iso, C15:0 ante, C 15:0, C10:0 iso, C17:0 iso, C16:1 and C17:0 ante, C17:0 and C17:1 are not presented in the table because no differences due to DS, LS or DSxLS ($P \geq 0.12$), and we did not hypothesize changes on those FA.

[2]PFAD = EnerGII as a source of palmitic and oleic acids (Virtus Nutrition LLC, Corcoran, Ca); EPA + DHA = StrataG113 as a source of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA; Virtus Nutrition LLC, Corcoran, Ca).

[3]*P*-values: DS = dam supplementation during the last 50 d of gestation; LS = lamb supplementation during the finishing period.

[4]SEM = most conservative SE for interaction was presented.

[5]CLA = conjugated linoleic acid; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; n-3 = omega-3; n-6 = omega-6.

[6]14:1 desaturase index = *cis*-9 C14:1/(C14:0 + *cis*-9 C14:1); 18:1 desaturase index = *cis*-9 C18:1/(C18:0 + *cis*-9 C18:1); CLA desaturase index = *cis*-9, *trans*-11 C18:2/(*cis*-9, *trans*-11 C18:2 + *trans*-11 C18:1.

compared with no oilseed supplementation. Thus, the results of the present and aforementioned studies suggest that marine FA sources may differentially affect the concentrations of FA with 16 or fewer carbons in subcutaneous adipose tissue compared with other sources of n-3 FA in ruminants. Omega-3 FA are typically thought to decrease lipogenic gene expression, leading to a potential decrease in FA synthesis (Clarke, 2001). Thus, the mechanism by which EPA and DHA in marine FA sources may potentially increase FA synthesis in subcutaneous adipose tissue is unknown but could be mediated by changes in gene expression. We have previously observed that lipogenic gene expression was increased in subcutaneous adipose tissue with supplementation of 0.39% (DM basis) of Ca salts containing EPA and DHA (Coleman et al., 2018a). However, as will be discussed, in the present study, there were few changes in the mRNA expression in subcutaneous adipose tissue with LS of EPA + DHA vs. PFAD. In addition, it is possible that the differences in FA synthesis could be related to biohydrogenation. Changes in tissue FA suggest that rumen biohydrogenation was altered with supplementation of Ca salts. It is possible that the Ca salt containing EPA and DHA was more highly biohydrogenated than the Ca salt of PFAD, which would have limited the amount of EPA and DHA being absorbed and reaching adipose tissue to exert the expected effects compared with palmitic and oleic acids.

Dam supplementation also altered the concentrations of FA with less than 16 carbons in lamb subcutaneous adipose tissue. Lambs born from ewes supplemented with EPA + DHA during late gestation had greater concentrations of C12:0 ($P = 0.01$) in their adipose tissue, while they tended to have greater concentrations of C10:0 ($P = 0.10$) than lambs born for PFAD ewes. To our knowledge, there are no other studies investigating the effects of maternal FA supplementation on lamb subcutaneous adipose tissue FA during the finishing period. As mentioned earlier, C10:0 and C12:0 may be synthesized in tissues. Thus, these results suggest that DS with EPA + DHA during late gestation may program lamb subcutaneous adipose tissue toward increased FA synthesis. However, as will be discussed in the gene expression section, the mechanism behind a potential increase in FA synthesis in adipose tissue with DS of EPA + DHA is unclear. Also, due to the design of the experiment we cannot confirm if these changes started during the fetal or growing stages.

Concentrations of C18:0 were greater in subcutaneous adipose tissue of lambs with LS of PFAD ($P = 0.04$). Fewer C18:1 isomers were affected by LS in adipose tissue compared with liver tissue. The concentration of C18:1 *cis*-11 tended ($P = 0.06$) to be greater in adipose tissue of lambs that were supplemented with EPA + DHA compared with PFAD. Concentrations of C18:1 *cis*-12, however, were greater in adipose tissue of lambs that were supplemented with PFAD during the finishing period ($P = 0.05$). The concentration of C18:2 *cis*-9, *trans*-11 was also greater ($P = 0.02$) in the adipose tissue of lambs supplemented with EPA + DHA. This contributed to the greater concentration of total CLA isomers in subcutaneous adipose tissue of EPA + DHA lambs ($P = 0.02$). Because the FA C18:0 is the end product of complete biohydrogenation in the rumen (Jenkins and Bridges, 2007), the greater concentration of C18:0 in subcutaneous adipose tissue of lambs supplemented with PFAD may be indicative of more complete biohydrogenation of the FA compared with EPA + DHA supplementation. This change in C18:0 with LS of EPA and DHA was also observed in muscle tissue, where C18:0 tended to be decreased with LS of EPA + DHA vs. PFAD (Carranza-Martin et al., 2018). While not significant, a numerical decrease in C18:0 ($P = 0.21$) was also observed in liver tissue with LS of EPA + DHA in the present study. The response of total CLA isomers in subcutaneous adipose tissue is opposite of liver tissue, where the concentration of total CLA isomers was greater with LS of PFAD compared with EPA + DHA. The reason for the different response between the 2 tissues is not known but may be attributed to a difference in storage vs. metabolism between adipose tissue and liver tissue.

Only one C18:1 isomer was significantly affected by DS in subcutaneous adipose tissue, with concentrations of C18:1 *cis*-9 being greater in lambs that were born from ewes supplemented with EPA + DHA vs. lambs from ewes supplemented with PFAD ($P = 0.01$). In addition, the C18:1 desaturase index was greater in subcutaneous adipose tissue when dams were supplemented with EPA + DHA during late gestation ($P = 0.05$). Thus, our results suggest that supplementing ewes with EPA + DHA during late gestation may increase the synthesis of C18:1 *cis*-9 in lamb subcutaneous adipose tissue during the finishing period. This is in line with the potential increase in FA synthesis discussed earlier for FA with less than 16 carbons. In addition, the

lack of changes in other C18:1 isomers due to DS is not unexpected since maternal supplementation during late gestation alone would have little influence on offspring rumen biohydrogenation pathways as described earlier.

A DS × LS interaction ($P \leq 0.04$) was observed for EPA and DHA concentration, where EPA + DHA supplementation of lambs during the finishing period increased the concentrations of EPA and DHA in subcutaneous adipose tissue, but the increase was greatest for lambs born from PFAD supplemented ewes vs. EPA + DHA supplemented ewes (Table 4). This same DS × LS interaction ($P \leq 0.04$) was observed for the concentration of total n-3 FA, and the n-6:n-3 FA ratio. This interaction between DS and LS is opposite of what we hypothesized. It is possible that the lambs with DS and LS of EPA + DHA were not synthesizing as much EPA and DHA in adipose tissue compared with the other lambs since they were receiving a steady supply of these FA. As will be described in the gene expression section, this assumption is supported by changes in mRNA expression. Although EPA and DHA were enriched in subcutaneous adipose tissue in the present study, concentrations of these FA were much lower in this depot compared with liver tissue. The concentrations of EPA and DHA in subcutaneous adipose tissue are also lower than the concentrations in muscle tissue of lambs in this study (Carranza-Martin et al., 2018). This is likely attributed to the fact that PUFA are preferentially incorporated into phospholipids in ruminants (Ashes et al., 1992) and phospholipids are more abundant in intramuscular fat. It is possible that EPA and DHA were preferentially incorporated into intramuscular fat, which may explain why concentrations of these 2 FA were greater in muscle tissue (Carranza-Martin et al., 2018). Liver tissue has also been characterized as having a greater concentration of phospholipids compared with other tissues in ruminants (Bermingham et al., 2018), which may explain why liver tissue had the greatest concentrations of EPA and DHA of the 3 tissues. However, it should be noted that the increases in EPA and DHA in liver and muscle tissue were only due to LS, and not DS (Carranza-Martin et al., 2018). This suggests that subcutaneous adipose tissue may have unique features that allowed an interaction of DS and LS to modulate concentrations of EPA and DHA. We speculate that the changes observed in adipose tissue with both DS and LS may be related to its function as the primary site of FA synthesis in ruminants (Bergen and Mersmann, 2005).

Concentration of mRNA in Liver Tissue

Fatty acid uptake and release genes in liver. There were no DS × LS interactions observed for the mRNA expression of genes related to FA uptake and release ($P > 0.10$; Table 5). Expression of *lipoprotein lipase* (***LPL***) was greater in liver tissue of lambs supplemented with PFAD during the finishing period ($P = 0.01$). Lipoprotein lipase is involved in the hydrolysis of FA from lipoproteins for uptake by cells. The change in *LPL* suggests that supplementation of EPA and DHA might decreased uptake of FA from lipoproteins by the liver compared with supplementation of palmitic and oleic acid. Lipoprotein lipase is a lipogenic enzyme due to its role in uptake of FA, thus the decrease in *LPL* expression with LS of EPA + DHA compared with PFAD fits with our hypothesis that lipogenic gene expression would be decreased by EPA and DHA supplementation. Data on the effects of supplementing n-3 FA on liver gene expression in ruminants are limited. Most studies in ruminants have examined the effects of supplementing sources of n-3 FA, such as EPA and DHA, on muscle and adipose tissue gene expression. However, in lactating dairy cows, hepatic *LPL* expression was not altered by supplementation with 200 g/d of fish oil or microalgae compared with no lipid supplementation (Vahmani et al., 2014). Another study in lactating dairy cows by Hiller et al. (2013) observed no difference in *LPL* when algae was supplemented with sunflower oil or linseed oil compared with SFA supplementation. The difference in the response of *LPL* in our study vs. the studies by Hiller et al. (2013) and Vahmani et al. (2014) could be attributed to differences in physiological status between lactating cows and growing lambs. The use of oil and algae, which may be biohydrogenated at a greater rate than Ca salts, may also be a factor in the differences between studies.

Dam supplementation altered hepatic *hormone-sensitive lipase* (***HSL***), with greater expression in lambs born from EPA + DHA ewes than lambs born from PFAD ewes ($P = 0.01$; Table 5). In addition, DS of EPA + DHA tended ($P = 0.09$) to increase the expression of fatty acid binding protein 4 (***FABP4***) in finishing lamb liver tissue compared with DS of PFAD. These changes in *HSL* and *FABP4* mRNA concentration suggest that maternal supplementation of EPA and DHA may program lamb liver tissue toward greater lipolysis. The fact that both of these were altered in the same direction supports the hypothesis that there is a direct interaction between FABP4 and HSL, where FABP4

Table 5. Relative mRNA expression in liver tissue of finishing lambs supplemented with Ca salts of EPA + DHA or palmitic fatty distillate acid (PFAD) at 1.48% and born from ewes supplemented with EPA + DHA or PFAD at 0.39% DM during the last 50 d of gestation[1]

Dam	PFAD		EPA + DHA			*P*-value[2]		
Lamb	PFAD	EPA + DHA	PFAD	EPA + DHA	SEM[4]	LS	DS	DS × LS
Item[3]								
5-LOX	19.98	15.16	17.87	16.38	1.88	0.04	0.77	0.25
ATGL	337.47	316.94	348.54	402.09	55.78	0.74	0.35	0.46
COX-2	35.65	47.74	49.13	43.26	8.16	0.64	0.52	0.19
Δ5-desaturase	2,186.21	1,010.13	2,172.50	1,173.61	218.95	<0.01	0.68	0.61
Δ6-desaturase	3,630.38	1,557.30	3,540.81	1,563.17	414.84	<0.01	0.91	0.90
DGAT1	391.60	338.52	471.02	466.36	69.55	0.64	0.11	0.69
DGAT2	664.13	756.79	750.12	748.65	93.87	0.59	0.64	0.57
ELOVL2	168.73	155.40	210.40	155.63	34.09	0.27	0.49	0.50
ELOVL4	17.27	14.64	17.26	22.28	3.01	0.63	0.14	0.13
ELOVL5	3,037.87	2,466.29	3,029.02	2,893.95	207.59	0.06	0.27	0.24
FABP4	14.32	15.02	22.24	23.29	5.17	0.85	0.09	0.97
FAS	506.94	248.18	563.97	320.86	71.04	<0.01	0.31	0.90
FATP1	19.58	22.00	24.21	22.17	4.20	0.96	0.52	0.55
HSL	12.23	13.07	18.18	16.53	1.94	0.82	0.01	0.46
LPL	48.32	40.37	59.30	37.22	6.77	0.01	0.46	0.17
SCD	3,673.38	1,373.02	3,464.32	1,170.47	225.78	<0.01	0.31	0.99

[1]*PFAD* = EnerGII as a source of palmitic and oleic acids (Virtus Nutrition LLC, Corcoran, Ca); EPA + DHA = StrataG113 as a source of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA; Virtus Nutrition LLC, Corcoran, Ca).

[2]*P*-values: DS = Dam supplementation during the last 50 d of gestation; LS = Lamb supplementation during the finishing period.

[3]*ATGL = adipose triglyceride lipase*; *COX-2 = cyclooxygenase-2*; *DGAT1 = diacylglycerol acyltransferase 1*; *DGAT2 = diacylglycerol acyltransferase 2*; *ELOVL2 = elongation of very long chain fatty acid 2*; *ELVL4 = elongation of very long chain fatty acid 4*; *ELOVL5 = elongation of very long chain fatty acid 5*; *FABP4 = fatty acid binding protein 4*; *FAS = fatty acid synthase*; *FATP1 = fatty acid transport protein 1*; *HSL = hormone-sensitive lipase*; *LPL = lipoprotein lipase*; *SCD = stearoyl-CoA desaturase*. Concentration of glucose-dependent insulinotropic polypeptide receptor, insulin receptor, ghrelin receptor, peroxisome proliferator-activated receptors α, β/δ, and γ, retinoid × receptor are not presented in the table because no differences due to DS, LS, or DS × LS ($P \geq 0.15$).

[4]SEM = most conservative SE for interaction was presented.

may facilitate lipolysis (Hotamisligil and Bernlohr, 2015). However, there were no differences between DS treatments in plasma NEFA concentrations to indicate that FA mobilization was increased by DS of EPA + DHA (Carranza-Martin et al., 2018).

Fatty acid synthesis genes in liver. No DS × LS interactions were observed for genes related to FA synthesis in liver tissue ($P > 0.10$; Table 5). Hepatic expression of *fatty acid synthase* (***FAS***) was lower ($P < 0.01$; Table 5) with LS of EPA + DHA. Lambs supplemented with PFAD during the finishing period had greater ($P < 0.01$) mRNA expression of *SCD*, *Δ5-desaturase*, and *Δ6-desaturase* compared with lambs supplemented with EPA + DHA. *Elongation of very long chain fatty acid 5* (*ELOVL5*) also tended to be greater ($P = 0.06$) in liver tissue of lambs with LS of PFAD. No differences were observed in the expression of other lipogenic genes in liver tissue between LS or DS ($P > 0.10$; Table 5): *elongation of very long chain fatty acid 2* (***ELOVL2***), *elongation of very long chain fatty acid 4* (***ELOVL4***), *diacylglycerol acyltransferase 1* (***DGAT1***), and *diacylglycerol acyltransferase 2* (***DGAT2***). Fatty acid synthase is used to synthesize SFA up to 16 carbons in length. The elongation enzyme ELOVL5 is utilized to produce longer chain FA that are typically polyunsaturated (Tvrdik et al., 2000). As mentioned earlier, the enzyme SCD introduces a double bond at the ninth carbon from the carboxyl end of FA with 12 to 19 carbons, whereas the Δ^5- and Δ^6-desaturases add double bonds at the fifth and sixth carbons, respectively (Nakamura and Nara, 2004). Therefore, the decreased expression of *FAS*, *SCD*, *Δ^5-desaturase*, *Δ^6-desaturase*, and *ELOVL5* in liver tissue with LS of EPA + DHA suggests FA synthesis was potentially decreased compared with LS of PFAD. This is supported in part by the changes in medium chain FA and desaturase indices for SCD in liver tissue with LS of EPA + DHA as described earlier. The alterations in lipogenic genes observed in the present study are consistent with previous studies. Hiller et al. (2013) supplemented dairy cows with a saturated rumen-protected fat, sunflower oil with algae, and linseed oil with algae and observed decreases in the liver tissue expression

of *FAS*, *SCD*, *Δ⁵-desaturase*, and *Δ⁶-desaturase* when the oils with algae were fed. In another study with dairy cattle, supplementation of fish oil or microalgae decreased expression of *FAS*, *SCD*, *Δ⁵-desaturase*, *Δ⁶-desaturase* in liver tissue compared with no fat supplementation (Vahmani et al., 2014). In dairy goats, supplementation of sunflower oil and fish oil together decreased expression of *SCD* and *Δ⁶-desaturase* in liver tissue compared with sunflower oil alone (Toral et al., 2013). Care should be taken in comparing the present study to the aforementioned studies due to the differences in the sources of EPA and DHA. However, it seems that supplementing a source of EPA and DHA has the expected effects of decreasing lipogenic gene expression in liver tissue across ruminant species.

There were no differences ($P \geq 0.11$; Table 5) between DS treatment for any of the measured genes in liver tissue that are involved in FA synthesis. Although not significant, the mRNA expression of these genes, except for *SCD*, was numerically greater in liver tissue of lambs born from ewes supplemented with EPA + DHA during late gestation. This is opposite of what we hypothesized, however, a similar effect of EPA and DHA on increasing expression of lipogenic genes was observed in subcutaneous adipose tissue of the dams of the lambs for this study (Coleman et al., 2018a). The dose of EPA and DHA supplemented to the dams was low at 0.39% DM and the concentrations of EPA and DHA in the lamb's plasma at birth were not different between lambs born from EPA + DHA supplemented ewes and those born from PFAD supplemented ewes (Coleman et al., 2018a). Thus, it is possible that this dose was not high enough to result in large amounts of EPA and DHA transfer and deposition to exert the expected effects via PPARα in liver tissue during fetal development.

Transcription factor genes in liver. Hepatic expression of all 3 *PPAR* isoforms, *α*, *γ* and *β*/*δ*, and the *retinoid × receptor alpha* (***RXR***α) was not different between DS during late gestation ($P \geq 0.77$), or LS during the finishing period ($P \geq 0.15$) and there were no DS × LS interactions ($P \geq 0.32$). The lack of changes in genes for transcription factors is likely due to the fact that these transcription factors are not highly transcriptionally regulated. However, long-chain FA have been observed to alter expression of *PPAR* isoforms in ruminant liver tissue in vitro (Bionaz et al., 2013). Thus, we hypothesized that PPARα expression and activation would be increased by supplementation with EPA and DHA in maternal and finishing diets. The lack of changes in the mRNA expression of genes for transcription factors agrees with previous in vivo studies in ruminants. The expression of *PPARγ* and *PPARα* in liver tissue of dairy goats was unaffected by supplementation of fish oil with sunflower oil vs. supplementation of sunflower oil alone (Toral et al., 2013). Vahmani et al. (2014) also observed no difference in the expression of *PPARγ* in liver tissue of lactating dairy cows supplemented with fish oil or microalgae compared with a nonfat control. Another study in lactating dairy cattle by Hiller et al. (2013) also observed no difference in hepatic *PPARγ* expression when sunflower oil with algae, linseed oil with algae, or a rumen-protected saturated fat were fed.

Although the expression of *PPARα* in liver tissue of finishing lambs in the present study was not altered by LS, it should be noted that there is indication that PPARα activation was increased. This is indicated by the changes observed in *FAS*, *SCD*, *Δ⁵-desaturase*, *Δ⁶-desaturase*, *LPL*, and *ELOVL5* in liver tissue with LS of EPA + DHA vs. PFAD, which are genes that have been identified as potential targets of PPAR in ruminants (Bionaz et al., 2013). There is only limited indication that DS may have also increased PPARα activation in offspring liver because *HSL* alone was altered with DS of EPA + DHA vs. PFAD. Thus, it seems that finishing diet supplementation of EPA and DHA may have had a greater effect on PPARα activation compared with maternal supplementation. As mentioned earlier, it is possible that the low dose of 0.39% DM of Ca salts of EPA and DHA used in the maternal diets was too small to illicit the expected effects on PPARα activation in offspring liver tissue.

Hormone receptor genes in liver. There were also no differences ($P \geq 0.16$) in the expression of *ghrelin receptor*, *insulin receptor*, or *glucose-dependent insulinotropic polypeptide* (**GIP**) *receptor* in liver tissue between LS or DS. The lack of differences in the expression of the hormone receptors in liver tissue was associated with the lack of change in plasma glucose and NEFA concentration with DS or LS reported in the companion article (Carranza-Martin et al., 2018). However, supplementation with increasing doses of Ca salts containing EPA and DHA to pregnant ewes during late gestation increased plasma glucose and decreased plasma ghrelin concentration (Nickles et al., 2018).

Inflammatory response genes in liver. The expression of *5-lipoxygenase* (***5-LOX***) was decreased (LS: $P = 0.04$) in liver tissue of lambs supplemented

with EPA + DHA compared with PFAD during the finishing period. The enzyme 5-LOX functions in the production of leukotrienes from FA. Both EPA and DHA may potentially reduce the expression of inflammatory response genes, such as *5-LOX* via signaling through transcription factors, such as PPAR, which may inhibit nuclear factor kappa B (**NF-kB**; Lee et al., 2010). The nuclear factor NF-kB signals cytokine that often produces inflammatory responses (Sordillo, 2016). Therefore, the decrease in mRNA expression of *5-LOX* with LS of EPA + DHA compared with PFAD is potentially beneficial at reducing the severity of an inflammatory response in the liver.

There were no differences ($P \geq 0.37$; Table 5) in the mRNA expression of *5-LOX* or *cyclooxygenase-2* (***COX-2***) in liver tissue between DS treatments. The lack of differences in the expression of these genes in liver tissue due to maternal FA supplementation could be related to the fact the dose of EPA and DHA was too low to result in changes in the expression of these in liver tissue or that DS does not modulate the expression of these genes in liver tissue. Even though the targeted dose of 18 mg/kg metabolic BW was the same for both ewes and lambs, the lambs were directly consuming the FA during the finishing period, rather than receiving them through placental transfer. The direct consumption during the finishing period could explain why changes were observed in liver tissue expression of inflammatory response genes with LS and not DS. In addition, to our knowledge, the ewes were not under inflammatory stress during the experiment and their plasma concentrations of prostaglandins were not different between treatments (Coleman et al., 2018a). Thus, the fetus was not exposed to an inflammatory environment that would potentially alter and program liver mRNA expression of inflammatory response genes.

Concentration of mRNA in Subcutaneous Adipose Tissue

Fatty acid uptake and release genes in subcutaneous adipose tissue. A tendency for a DS × LS interaction ($P = 0.08$; Table 6) was observed for *HSL* in subcutaneous adipose tissue where lambs with DS and LS of EPA + DHA had the lowest expression of *HSL* compared with the other dam and lamb treatments. Due to the role of HSL in triglyceride breakdown, the DS × LS interaction observed for *HSL* expression in subcutaneous adipose tissue might suggest that lipolysis was decreased in lambs with DS and LS of EPA + DHA. This interaction is opposite of what we hypothesized would occur; however, this change fits with gene expression changes in the dam's subcutaneous adipose tissue; dams supplemented with EPA + DHA had increased expression of lipogenic genes (Coleman et al., 2018a). The changes in expression of *HSL* between adipose and liver tissue seem contradictory because the mRNA concentration of *HSL* increased with DS of EPA + DHA compared with PFAD. However, the expression of *HSL* was much greater in subcutaneous adipose tissue compared with liver tissue, suggesting that the function of the gene is tissue specific, which may be why the response is different between the 2 tissues.

Fatty acid synthesis genes in subcutaneous adipose tissue. A DS × LS interaction ($P = 0.01$; Table 6) was observed for *ELOVL2* where lambs that were born from DS of EPA + DHA and LS of EPA + DHA had the lowest expression of *ELOVL2* in adipose tissue. The enzyme ELOVL2 may be used in the synthesis of EPA and is utilized to synthesize DHA (Guillou et al., 2010). Thus, changes in *ELOVL2* mRNA expression suggest that synthesis of EPA and DHA may have been decreased in lambs with DS and LS of EPA + DHA. It is possible that the expression of *ELVOL2* was downregulated to limit the production of EPA and DHA due to the steady supply of these FA both prenatally and during the finishing period. This potential decrease in EPA and DHA synthesis may also explain why lambs with DS and LS of EPA + DHA did not have the greatest concentrations of EPA and DHA in their adipose tissue compared with the other DS and LS combinations. The lack of LS effects on genes for FA synthesis contrasts with the effects observed in the dam's subcutaneous adipose tissue when they were supplemented with the same dose of EPA and DHA at 18 mg/kg of metabolic BW; dams supplemented with EPA + DHA had increased expression of *FAS* and *DGAT2*, potentially increasing lipogenesis (Coleman et al., 2018a). In addition, the lack of LS or DS effects on the expression of genes for FA synthesis does not explain the mechanism by which synthesis of medium chain FA in subcutaneous adipose may have been increased by of EPA + DHA vs. PFAD. It is possible that protein expression was altered, even if mRNA was not, to result in changes in FA synthesis. However, protein expression was not measured in this study to support this assumption. In addition, compared with liver tissue, there were fewer changes in the mRNA expression of genes related to FA synthesis in subcutaneous adipose tissue. This difference in expression between

Table 6. Relative mRNA expression in subcutaneous adipose tissue of finishing lambs supplemented with Ca salts of EPA + DHA or palmitic fatty distillate acid (PFAD) at 1.48% and born from ewes supplemented with EPA + DHA or PFAD at 0.39% DM during the last 50 d of gestation[1]

Dam	PFAD		EPA + DHA			*P*-value[2]		
Lamb	PFAD	EPA + DHA	PFAD	EPA + DHA	SEM[3]	LS	DS	Ds × LS
Item[4]								
5-LOX	16.37	19.28	13.65	14.96	4.194	0.58	0.37	0.84
ATGL	1,628.63	1,324.04	1,195.24	1,664.91	290.39	0.76	0.86	0.16
Adiponectin	97,006	91,368	116,823	81,843	12,424	0.07	0.64	0.20
COX-2	118.38	193.06	225.68	131.82	86.97	0.90	0.77	0.30
Δ5-desaturase	196.49	200.31	197.51	175.50	27.47	0.72	0.64	0.61
Δ6-desaturase	121.82	129.67	132.15	113.53	20.25	0.77	0.88	0.48
DGAT1	569.16	585.64	595.82	824.43	185.58	0.46	0.44	0.53
DGAT2	12,682	12,404	15,900	12,369	2277	0.37	0.45	0.44
ELOVL2	30.50	35.75	35.88	27.42	3.043	0.51	0.55	0.01
ELOVL4	63.70	60.01	65.35	55.41	5.330	0.17	0.76	0.53
ELOVL5	2015.85	2,253.16	2,213.47	1,703.84	321.37	0.64	0.52	0.21
FABP4	237,594	229,692	252,470	195,327	22,119	0.12	0.63	0.23
FAS	35,407	29,439	32,912	32,719	6,282	0.59	0.95	0.62
FATP1	27.22	26.97	21.04	25.42	5.67	0.70	0.47	0.67
Ghrelin receptor	19.92	13.92	17.00	26.89	5.454	0.70	0.32	0.12
GIP receptor	67.98	66.99	62.78	56.04	3.897	0.20	0.01	0.34
HSL	6,115.46	6,345.70	7,395.30	4,725.23	869.75	0.14	0.83	0.08
Insulin receptor	469.21	483.81	439.52	430.10	48.86	0.95	0.36	0.79
Leptin	477.09	395.68	574.91	366.52	85.48	0.08	0.66	0.42
LPL	35,794	36,538	40,834	33,865	5,337	0.53	0.81	0.44
PPARα	213.28	201.01	211.47	198.53	16.89	0.42	0.89	0.98
PPARβ/δ	125.38	113.35	110.51	140.74	17.72	0.58	0.70	0.20
PPARγ	4,090.70	4,371.56	4,565.76	4,045.50	389.57	0.72	0.83	0.26
Resistin	2.78	4.92	2.91	4.20	1.15	0.07	0.75	0.65
RXRα	114.91	111.57	121.19	130.56	17.39	0.85	0.43	0.69
SCD	17,213	19,746	22,287	16,138	4,438	0.65	0.86	0.29

[1]*PFAD* = EnerGII as a source of palmitic and oleic acids (Virtus Nutrition LLC, Corcoran, Ca); EPA + DHA = StrataG113 as a source of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA; Virtus Nutrition LLC, Corcoran, Ca).

[2]*P*-values: DS = Dam supplementation during the last 50 d of gestation; LS = Lamb supplementation during the finishing period.

[3]SEM = most conservative SE for interaction was presented.

[4]*5-LOX = 5-lipoxygenase*; *ATGL = adipose triglyceride lipase*; *COX-2 = cyclooxygenase-2*; *DGAT1 = diacylglycerol acyltransferase 1*; *DGAT2 = diacylglycerol acyltransferase 2*; *ELOVL2 = elongation of very long chain fatty acid 2*; *ELVL4 = elongation of very long chain fatty acid 4*; *ELOVL5 = elongation of very long chain fatty acid 5*; *FABP4 = fatty acid binding protein 4*; *FAS = fatty acid synthase*; *FATP1 = fatty acid transport protein 1*; *GIP = glucose-dependent insulinotropic polypeptide*; *HSL = hormone-sensitive lipase*; *LPL = lipoprotein lipase*; *PPAR = peroxisome proliferator-activated receptor*; *RXR = retinoid × receptor*; *SCD = stearoyl-CoA desaturase*.

tissues could be associated with the greater metabolic activity of the liver and its role in lipid metabolism, compared with the role of adipose tissue in lipid storage and synthesis.

Transcription factor genes in subcutaneous adipose tissue. As in liver tissue, expression of all 3 *PPAR* isoforms *α*, *γ* and *β*/*δ*, and *RXRα* was not different ($P \geq 0.20$; Table 6) in subcutaneous adipose tissue between DS or LS and there were no DS × LS interactions. In accordance with the present study, we also observed no differences in the expression of genes for transcription factors in the subcutaneous adipose tissue of the dams used for this study (Coleman et al., 2018a). We hypothesized that PPARα activation and expression could be increased by supplementation with EPA and DHA and would increase the expression of lipolytic genes and decrease the expression of lipogenic genes. Although there is evidence of PPARα activation in liver tissue in the present study via LS and DS, there were few changes in subcutaneous adipose tissue gene expression to suggest that PPARα could be activated there. The lack of differences in lipolytic gene expression in subcutaneous adipose tissue in the present study could be related to differences in the expression of PPARs across tissues, as *PPARα* was greatest in liver and *PPARγ*

was greatest in adipose tissue. In addition, much of the basis for what is known about PPARα is based on effects in liver tissue (Clarke, 2001). Although there is also evidence from rodent models (Flachs et al., 2005), as well as in vitro models with adipocytes (Guo et al., 2005), that EPA and DHA may increase the potential for FA oxidation in adipose via gene expression changes, it is possible that effects on gene expression may not be as potent in adipose tissue compared with liver. The effects of PPARγ may be more potent in adipose tissue due to its effects on lipid storage (Nakamura et al., 2014). Even though both DS and LS increased deposition of EPA and DHA in subcutaneous adipose tissue, it is possible that our dose of 18 mg/kg metabolic BW was too low to activate PPARα there. Another factor for the lack of effects on lipolytic gene expression could be free fatty acid receptor 4 (FFAR4). Both EPA and DHA can serve as ligands of FFAR4, which is expressed in adipose, and has effects on adipogenesis and inflammation (Oh and Walenta, 2014). Free fatty acid receptor 4 may have a proadipogenic function in adipose tissue, as findings in humans and mice suggest that dysfunction of FFAR4 may decrease adipocyte differentiation and lipogenesis and enhance hepatic lipogenesis in obesity (Ichimura et al., 2012). In addition, the knockdown of FFAR4 in 2T3-L1 murine adipocytes inhibited the expression of adipogenic genes and impaired lipid accumulation (Gotoh et al., 2007). Thus, binding of ligands to FFAR4 may promote expression of lipogenic genes. Expression of *FFAR4* in bovine subcutaneous adipose tissue has been confirmed by Agrawal et al. (2017). Although the expression of *FFAR4* was not measured in the present study, it is possible that EPA and DHA could have been bound by FFAR4 in subcutaneous adipose tissue to increase lipogenic gene expression. This possible binding may explain the potential for increased FA synthesis observed in subcutaneous adipose tissue with LS and DS of EPA + DHA compared with PFAD.

Adipokine and hormone receptor genes in subcutaneous adipose tissue. In subcutaneous adipose tissue, there was a tendency for the expression of both *adiponectin* (P = 0.07; Table 6) and *leptin* (P = 0.08) to be lower in lambs supplemented with EPA + DHA compared with PFAD. The expression of *resistin* in adipose tissue also tended (P = 0.07) to be greater with LS of EPA + DHA. Leptin is an adipokine that has effects on food intake and increasing energy expenditure (Contreras et al., 2017), whereas adiponectin improves insulin sensitivity and lipogenesis in adipose tissue (Stern et al., 2016). Adiponectin and leptin have been shown to have opposite concentrations (Matsubara et al., 2002), so it is surprising that there was a tendency for adiponectin and leptin to both decrease with LS of EPA + DHA vs. PFAD. Little is known about the functions of resistin across species; however, studies in dairy cattle have found that resistin is positively correlated with NEFA concentrations, suggesting a role in increasing lipolysis (Reverchon et al., 2014; Weber et al., 2016). As presented in a companion paper, no differences were observed in the plasma concentration of NEFA in our lambs (Carranza-Martin et al., 2018). Thus, the tendency for higher *resistin* expression in subcutaneous adipose tissue with LS of EPA + DHA compared with PFAD does not fit with our data for plasma metabolites. It is possible that the expression of genes for adipokines did not translate to changes in protein expression. This may explain why the mRNA expression of *adiponectin* and *leptin* were not opposite as expected, and why the expression of *resistin* does not fit with plasma NEFA concentrations. However, as mentioned earlier, protein expression, nor plasma concentrations of adipokines, were measured in this study. The lack of differences in the mRNA expression of *GIP receptor*, *insulin receptor*, and *ghrelin receptor* is not unexpected because, as mentioned earlier, plasma concentrations of metabolites were not different between LS of EPA + DHA and PFAD (Carranza-Martin et al., 2018).

Only one DS effect was observed for the adipokine and hormone receptor genes measured in subcutaneous adipose tissue; expression of the *GIP receptor* in adipose tissue was lower in lambs born from dams supplemented with EPA + DHA vs. PFAD during late gestation (P = 0.01). The DS effect on mRNA expression of *GIP receptor* is unexpected due to the lack of DS effects on plasma metabolites in the present study (Carranza-Martin et al., 2018). The GIP receptor is expressed in adipocytes, where GIP stimulation increases the uptake of nutrients into adipocytes (Yamada and Seino, 2004). Thus, without differences in plasma metabolites, the lambs would not have needed increased GIP receptors to take up more nutrients. In addition, there were no differences in the plasma concentrations of insulin and ghrelin, or metabolites of their dams (Coleman et al., 2018b), or in the expression of genes for hormone receptors in the subcutaneous adipose tissue of their dams (Coleman et al., 2018a). Without changes in metabolites or maternal hormone receptors in adipose tissue, it

is unclear why the expression of *GIP receptor* was different in finishing lamb subcutaneous adipose tissue with DS of EPA + DHA compared with PFAD. However, Miyawaki et al. (2002) showed that GIP and its receptor are associated with feed efficiency, where GIP receptor knockout mice had a lower respiratory quotient when fed a high-energy diet, indicating that they were using more fat as an energy source. We report in the companion paper (Carranza-Martin et al., 2018) that lambs with DS of EPA + DHA were, on average, heavier during the finishing period than lambs with DS of PFAD. This difference in BW could be associated with the decreased expression of *GIP receptor* in the subcutaneous adipose tissue, independent of the metabolite or hormone concentrations. More work is needed to understand the potential associations between maternal supplementation of EPA and DHA and offspring performance and *GIP receptor* mRNA expression.

Inflammatory response genes in subcutaneous adipose tissue. No differences ($P > 0.30$; Table 6) were observed between DS or LS in the expression of *COX-2* or *5-LOX* in subcutaneous adipose. The expression of inflammatory response genes in subcutaneous adipose tissue is different than that of liver tissue, where *5-LOX* was altered by LS. As neither tissue was in an inflammatory state, it is not known why expression of *5-LOX* was altered in liver and not subcutaneous adipose tissue. As described earlier, EPA and DHA may modulate expression of *5-LOX* via PPAR (Lee et al., 2010). Compared with subcutaneous adipose, changes in hepatic expression of genes related to FA synthesis, uptake, and release suggest a greater activation of PPARα by LS of EPA + DHA vs. PFAD. This greater activation in liver tissue may explain why *5-LOX* expression was altered in liver vs. subcutaneous adipose tissue, even though the level of expression was similar between the 2 tissues.

An interaction exists between DS and LS of Ca salts enriched with EPA and DHA on lamb tissue FA concentrations and mRNA expression. However, the response may be tissue dependent as more interactions between DS and LS were observed in subcutaneous adipose tissue compared with liver tissue. In addition, dam and lamb supplementation with an enriched source of EPA and DHA both independently altered liver and subcutaneous adipose tissue FA and mRNA expression. Importantly, supplementation with EPA and DHA during the finishing period increased the concentrations of these FA in both liver and subcutaneous adipose tissue. Lamb supplementation of an enriched source of EPA and DHA altered hepatic FA and mRNA concentrations toward increased lipolysis and decreased lipogenesis compared with supplementation of a Ca salt of PFAD. Maternal supplementation of Ca salts enriched with EPA and DHA during late gestation may alter liver tissue in the same way; however, changes were less evident. Changes in subcutaneous adipose tissue FA suggest that maternal and finishing period supplementation with EPA and DHA altered lipogenesis. However, there were no changes in mRNA expression of lipogenic genes with DS or LS of an enriched source of EPA and DHA compared with a source of PFAD. Thus, more work is needed to understand the mechanisms by which supplementing Ca salts enriched in EPA and DHA to dams during late gestation and their offspring during the finishing period may have increased lipogenesis, and the potential fetal programming effects of these FA.

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