Effects of supplementing calcium salts of polyunsaturated fatty acids to late-gestating beef cows on performance and physiological responses of the offspring¹

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ABSTRACT: This experiment compared performance and physiological responses of the offspring from cows supplemented with Ca salts of PUFA or SFA + MUFA during late gestation. Ninety-six multiparous, nonlactating, pregnant Angus × Hereford cows were ranked by BW, BCS, and age and divided into 24 groups of 4 cows/group at the end of their second trimester of gestation (d -7). Cows conceived during the same estrus synchronization + AI protocol, with semen from a single sire; hence, gestation length was 195 d for all cows at the beginning of the experiment (d 0). Groups were randomly assigned to receive (DM basis) 405 g/cow daily of soybean meal in addition to 1) 190 g/cow daily of Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids or 2) 190 g/cow daily of Ca salts of SFA + MUFA based on palmitic and oleic acids (CON). Groups were maintained in 2 pastures (6 groups of each treatment/pasture) and received daily 10.1 kg/cow (DM basis) of grass-alfalfa hay. Groups were segregated into 1 of 12 drylot pens (6 by 18 m) and individually offered treatments 3 times/wk from d 0 until calving. Cow BW and BCS were recorded, and blood samples were collected on d-7 of the experiment and also within 12 h after calving. Calf BW was also recorded within 12 h of calving. Calves were weaned on d 280 of the experiment, preconditioned for 45 d (d 280 to 325), transferred to a growing lot on d 325, and moved to a finishing lot on d 445, where they remained until slaughter. At calving, PUFA-supplemented cows had a greater (P < 0.01) proportion (as % of total plasma fatty acids) of PUFA, including linoleic, linolenic, arachidonic, docosapentaenoic, and docosahexaenoic acids. At weaning, calves from CON-supplemented cows were older (P = 0.03), although no treatment differences were detected (P = 0.82) for calf weaning BW. During both growing and finishing phases, ADG was greater $(P \le 0.06)$ in calves from PUFA-supplemented cows. Upon slaughter, HCW and marbling were also greater $(P \le 0.05)$ in calves from PUFA-supplemented cows. Collectively, these results indicate that supplementing eicosapentaenoic, docosahexaenoic, and linoleic acids to late-gestating beef cows stimulated programming effects on postnatal offspring growth and carcass quality. Therefore, supplementing late-gestating beef cows with Ca salts of PUFA appears to optimize offspring productivity in beef production systems.

Key words: beef cows, offspring, polyunsaturated fatty acids, pregnancy, supplementation

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INTRODUCTION

Maternal nutrition is a major extrinsic factor programming nutrient partitioning and development of fetal organ systems associated with health, production, and reproduction (Long et al., 2010; Silvestre et al., 2011; Garcia et al., 2014a). Hence, nutritional management of late-gestating beef cows has been shown to directly impact performance of the subsequent offspring through fetal programming effects (Funston et al., 2010; Bohnert et al., 2013; Marques et al., 2016b). However, the majority of research conducted to date within this subject focused on energy and CP nutrition, and little is known about the potential impacts of supplementing PUFA to gestating cows on offspring productivity.

In humans and livestock species, *n*-3 and *n*-6 PUFA play critical roles in several body functions. However, PUFA such as linoleic acid cannot be synthesized by the body and must be consumed through the diet (Hess et al., 2008). During gestation, dietary PUFA becomes available in the circulation and are transferred to the fetus via the placenta (Noble et al., 1978; Garcia et al., 2014b). In humans, supplementing pregnant women with PUFA is considered critical for optimal fetal and early-life child development, including growth, nervous, and immune responses (Greenberg et al., 2008). Accordingly, research with swine reported that supplementing pregnant sows with PUFA benefited piglet vitality as well as pre- and postweaning growth (Tanghe and De Smet, 2013).

Based on these researches, we hypothesized that supplementing PUFA to late-gestating beef cows will increase postnatal offspring productivity. Nevertheless, PUFA should be supplemented to cattle and other ruminant livestock as rumen-protected sources, such as Ca salts, to prevent extensive ruminal biohydrogenation (Sukhija and Palmquist, 1990). Hence, this experiment evaluated the effects of supplementing Ca salts of PUFA to beef cows during the last trimester of gestation on performance and physiological responses of the offspring.

MATERIALS AND METHODS

This experiment was conducted at the Oregon State University — Eastern Oregon Agricultural Research Center (Burns station; Burns, OR). The animals used were cared for in accordance with acceptable practices and experimental protocols reviewed and approved by the Oregon State University, Institutional Animal Care and Use Committee (number 4758).

Cow-Calf Management and Dietary Treatments

Ninety-six multiparous, nonlactating, pregnant Angus \times Hereford cows (586 \pm 4 kg BW, 7.5 \pm 0.2 yr of

age, and BCS of 5.01 ± 0.03 according to Wagner et al. [1988]) were assigned to this experiment at the end of their second trimester of gestation. Cows conceived during the same estrus synchronization + AI protocol using semen from a single Angus sire, according to the breeding management and pregnancy diagnosis described by Cooke et al. (2014). At the beginning of the experiment (d 0), gestation length was 195 d for all cows.

Prior to the beginning of the experiment (d-7), cows were ranked by BW, BCS, and age and divided into 24 groups of 4 cows/group. Groups were then randomly assigned to receive (DM basis) 405 g of soybean meal per cow daily in addition to 1) 190 g/cow daily of Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids (95 g of Prequel + 95 g of Strata, equivalent to 100 g, as fed, of each product; Virtus Nutrition, LLC, Corcoran, CA) or 2) 190 g/cow daily of Ca salts of SFA + MUFA based on palmitic and oleic acids (CON; 190 g of EnerGII, equivalent to 200 g, as fed, of this product; Virtus Nutrition, LLC). Supplement treatments were isonitrogenous, isolipidic, and isocaloric (Table 1). Groups were maintained in 1 of 2 meadow foxtail (Alopecurus pratensis L.) pastures (25-ha pastures, 12 groups/pasture, and 6 groups/treatment in each pasture) beginning on d -7. Grass-alfalfa hay was provided daily at 10.1 kg/ cow (DM basis), and cows had ad libitum access to water and a commercial mineral + vitamin mix (Cattleman's Choice; PerforMix Nutrition Systems, Nampa, ID) containing 14% Ca, 10% P, 16% NaCl, 1.5% Mg, 6,000 mg/ kg Zn, 3,200 mg/kg Cu, 65 mg/kg I, 900 mg/kg Mn, 140 mg/kg Se, 136 IU/g of vitamin A, 13 IU/g of vitamin D₃, and 0.05 IU/g of vitamin E. No forage was available for grazing due to previous hay harvest and snow cover resulting from wintery conditions. Cows remained in their respective groups from d -7 until calving.

From d 0 of the experiment until calving, cows were gathered 3 times weekly (Mondays, Wednesdays, and Fridays) and groups were sorted into 1 of 12 drylot pens (6 by 18 m). Groups were individually offered treatments (5.55 kg of supplement treatment/feeding per group, from 0800 to 1000 h, DM basis) in feed bunks with linear space of approximately 0.8 m/cow. Accordingly, Cook et al. (2017) reported that Ca salts of fatty acids can be offered to cows as infrequently as 3 times/wk without impairing fatty acid intake and subsequent circulating fatty acid concentrations. Groups promptly consumed treatments within 15 min after feeding. Cows were returned to the pasture immediately after their supplement was completely consumed. Grass-alfalfa hay was offered in feed bunks located in each pasture (linear space of approximately 1.0 m/cow). Diets (hay + treatments) were formulated to meet or exceed nutrient requirements for energy, CP, minerals, and vitamins (Table 1) of late-gestating beef cows (NRC, 2000).

Immediately after calving, cow—calf pairs were removed from their pasture and assigned to the general management of the research herd until weaning (described by Marques et al. [2016a]), which did not include PUFA or SFA + MUFA supplementation. Male calves were castrated at birth using an elastic castration band. All calves were administered Clostrishield 7 and Virashield 6 + Somnus (Novartis Animal Health, Bucyrus, KS) at approximately 30 d of age.

Calf Management

Preconditioning (d 280 to 325). Calves were weaned on d 280 of the experiment and transferred to a 6-ha meadow foxtail (Alopecurus pratensis L.) pasture, which had been previously harvested for hay, for a 45-d preconditioning period as a single group. Calves were administered One Shot Ultra 7, Bovi-Shield Gold 5, TSV-2, and Dectomax (Zoetis, Florham Park, NJ) at weaning, and received a booster of Bovi-Shield Gold 5, UltraChoice 7, and TSV-2 (Zoetis) 28 d after weaning (d 308 of the experiment). During preconditioning, calves received mixed alfalfa—grass hay (12% CP and 57% TDN, DM basis), water, and the same commercial mineral and vitamin mix previously described (Cattleman's Choice) for ad libitum consumption.

Growing (d 325 to 445) and Finishing (d 445 Until Slaughter). On d 325, all calves were loaded into a commercial livestock trailer and transported for 480 km to the growing lot (Top Cut, Echo, OR), where they remained for 120 d and were managed as a single group. On d 445, calves were moved to an adjacent finishing lot (Beef Northwest, Boardman, OR), where they continued to be managed as a single group until slaughtered at a commercial packing facility (Tyson Fresh Meats Inc., Pasco, WA). After arriving at the finishing lot, calves received Bovi-Shield Gold 5 (Zoetis), Vision 7 (Merck Animal Health, Kenilworth, NJ), Valbazen (Zoetis), and Bimectin pour-on (Bimeda Animal Health Inc., Oakbrook Terrace, IL). Steers were implanted with Revalor IS (Merck Animal Health) and heifers were implanted with Revalor IH (Merck Animal Health) on arrival. Growing and finishing diets, which did not contain Ca salts of PUFA or SFA + MUFA, were fed ad libitum (Table 2). Slaughter date was determined according to the availability of the commercial packing facility (Tyson Fresh Meats Inc.). As a result, calves were randomly assigned to slaughter on 2 separate dates, 11 d apart, regardless of treatment group (22 and 23 calves from CON- and PUFAsupplemented cows, respectively, after 121 d on feed [DOF] and 26 and 24 calves from CON- and PUFAsupplemented cows, respectively, after 132 DOF).

Table 1. Ingredient composition and nutrient profile of diets containing 190 g/cow daily of Ca salts of SFA + MUFA based on palmitic and oleic acids (CON) or 190 g/cow daily of Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids

Item	CON	PUFA
Ingredients, kg/d (DM basis)		
Grass–alfalfa hay	10.1	10.1
Soybean meal	0.405	0.405
EnerGII ¹	0.190	_
Prequel ¹	_	0.095
Strata ¹	_	0.095
Nutrient profile ² (DM basis)		
DM	93.5	93.5
TDN, ³ %	61	61
NEm, ⁴ Mcal/kg	1.29	1.28
CP, %	10.2	10.2
Fat, %	3.52	3.49
Palmitic acid (16:0), %	0.88	0.49
Stearic acid (18:0), %	0.10	0.13
Oleic acid (18:1), %	0.91	0.61
Linoleic acid (18:2), %	0.44	0.69
Linolenic acid (18:3), %	0.92	0.97
Eicosapentaenoic acid (20:5n-3), %	0.00	0.13
Docosahexaenoic acid (22:6n-3), %	0.00	0.11
Daily intake ⁵		
DM, kg	10.8	10.8
TDN, kg	6.6	6.6
NEm, Mcal	13.9	13.8
CP, kg	1.1	1.1
Fat, kg	0.381	0.377
Palmitic acid (16:0), g	94.7	53.2
Stearic acid (18:0), g	11.1	14.0
Oleic acid (18:1), g	98.7	65.4
Linoleic acid (18:2), g	47.9	74.6
Linolenic acid (18:3), g	99.0	104.7
Eicosapentaenoic acid (20:5n-3), g	0	14.5
Docosahexaenoic acid (22:6n-3), g	0	11.5

¹Calcium salts from Virtus Nutrition, LLC (Corcoran, CA).

Sampling

Feedstuffs. Two samples of all dietary ingredients fed to late-gestating cows (Table 3) were collected before the beginning of the experiment and analyzed for nutrient content by a commercial laboratory (Dairy One Forage Laboratory, Ithaca, NY). Each sample was analyzed in triplicate by wet chemistry procedures for concentrations of CP (method 984.13; Horwitz, 2006),

²Values obtained using wet chemistry analysis (Dairy One Forage Laboratory, Ithaca, NY). Fatty acid content was determined according to the procedures described by Moriel et al. (2015).

³Calculated according to the equations described by Weiss et al. (1992).

 $^{^4}$ Calculated with the following equation (NRC, 2000): NEm = 1.37 ME - 0.138 ME2 + 0.0105 ME3 - 1.12, given that ME = DE \times 0.82 and 1 kg of TDN = 4.4 Mcal DE.

⁵According to total intake and nutrient profile of dietary treatments.

Table 2. Ingredient composition (as-fed basis) of growing and finishing diets offered to cattle

Ingredients, % as fed	Grow	ing lot ¹	Finishing lot ²				
	A	В	A	В	С	D	Е
Alfalfa hay	0.0	0.0	23.3	16.7	8.4	6.6	6.6
Barley	18.0	17.0	0.0	0.0	0.0	0.0	0.0
Corn cobbs	0.0	5.3	0.0	0.0	0.0	0.0	0.0
Corn silage	10.0	15.0	0.0	0.0	0.0	0.0	0.0
Corn stover	0.0	10.0	0.0	0.0	0.0	0.0	0.0
Culled French fries	0.0	0.0	0.0	5.0	6.7	8.0	8.0
High-moisture corn	0.0	0.0	0.0	0.0	7.7	15.0	15.0
Mineral and vitamin mix ^{3,4}	3.0	3.4	11.3	7.2	6.5	3.0	3.0
Mixed pea/wheat/barley hay	34.0	5.3	0.0	0.0	0.0	0.0	0.0
Potato slurry	13.0	23.0	0.0	10.0	12.1	15.0	15.0
Rolled corn	0.0	0.0	40.4	40.0	40.0	36.0	36.0
Ryegrass silage	22.0	15.0	0.0	0.0	0.0	0.0	0.0
Vegetable oil	0.0	0.0	0.0	0.5	0.9	1.4	1.4
Wet distiller's grain	0.0	6.0	25.0	20.6	17.7	15.0	15.0

¹A = offered for 10 d on receiving; B = offered for 110 d after diet A and until transfer to the finishing lot.

ADF (method 973.18 modified for use in an Ankom 200 fiber analyzer [ANKOM Technology Corp., Fairport, NY]; Horwitz, 2006), and NDF (Van Soest et al., 1991; modified for use in an Ankom 200 fiber analyzer). Calculations for TDN used the equation proposed by Weiss et al. (1992), whereas NEm was calculated with the equations proposed by the NRC (2000). Feed samples were also analyzed for fatty acid profile, using the techniques described by Moriel et al. (2015).

Cows and Newborn Calves

Individual cow BW and BCS (Wagner et al., 1988) were recorded, and a blood sample was collected, via jugular venipuncture, prior to the beginning of the experiment (d –7; initial measurement). Within 12 h after calving, cow BW, cow BCS, calf birth BW, and calf gender were recorded and a blood sample was collected from each cow via jugular venipuncture.

Preconditioning. Cow BW and BCS (Wagner et al., 1988) were recorded at weaning (d 280). Calf BW was recorded and blood samples were collected on d 280, 282, 285, and 288 of the experiment via jugular venipuncture. During the 45 d of preconditioning, calves were observed daily for bovine respiratory disease (**BRD**) signs according to the subjective criteria described by Berry et al. (2004) and received 0.1 mL/kg of BW of Hexasol LA Solution (Norbrook Inc. USA, Overland Park, KS) when signs were observed. Preconditioning ADG was determined using BW obtained at weaning (d 280) and before shipping to the growing lot (d 325).

Growing and Finishing. Calf BW was recorded on arrival at the growing lot (d 325) and the finishing lot (d 425). Calves were observed daily for BRD signs according to the DART system (Zoetis) and received medication according to the management criteria of the growing and finishing yards. At the commercial packing plant, HCW was collected on slaughter. Final finishing BW was estimated based on HCW adjusted to a 63% dressing percentage (Loza et al., 2010). After a 24-h chill, trained personnel assessed carcass back fat thickness at the 12th-rib and LM area, whereas a USDA grader recorded all other carcass measures. Growing lot ADG was determined using BW values obtained on growing lot and finishing lot arrival (d 425). Finishing lot ADG was determined using BW values obtained on finishing lot arrival and final finishing BW estimated from HCW (Loza et al., 2010).

Blood Analysis

All blood samples were collected into commercial heparinized blood collection tubes (Vacutainer, 10 mL; Becton, Dickinson and Company, Franklin Lakes, NJ), centrifuged at $2,500 \times g$ for 30 min at 4°C for plasma collection, and stored at -80°C on the same day of collection.

Plasma samples from cows (d -7 of the experiment and on calving) were analyzed for fatty acid profile as previously described by Garcia et al. (2014a). Plasma samples collected from calves from d 280 to 288 were analyzed for haptoglobin (Cooke and Arthington, 2013) and cortisol (Immulite 1000; Siemens Medical Solutions

²A = offered for 10 d on receiving; B = offered for 10 d after diet A; C = offered for 10 d after diet B; D = offered for 30 d after diet C; E = offered until slaughter.

³Growing diets included Rumax (PerforMix Nutrition Systems, Nampa, ID).

⁴Finishing diets included a customized blend of minerals, vitamins, and feed additives (Westway Feed Products LLC, Tomball, TX, and Land O'Lakes, Inc., Saint Paul, MN).

Table 3. Nutritional and fatty acid profile of feedstuff offered to late-gestating beef cows

				Fat supplement ¹	
Item (DM basis)	Hay	Soybean meal	EnerGII	Prequel	Strata
DM, %	92	89	95	95	95
TDN, ² %	57	80	271	263	263
NEm, ² Mcal/kg	1.12	1.93	8.73	8.21	8.21
CP, %	8.80	49.80	0.00	0.00	0.00
Total fat, %	2.1	1.5	81.8	80.2	79.7
Fatty acid profile, % total fat ³					
Palmitic acid (16:0)	15.94	11.96	37.32	14.19	9.11
Palmitoleic acid (16:1)	0.00	0.00	0.00	0.00	12.93
Stearic acid (18:0)	4.03	5.08	1.35	2.59	3.84
Oleic acid (18:1)	7.94	10.61	50.5	33.07	27.76
Linoleic acid (18:2)	14.49	65.31	7.96	47.19	3.01
Linolenic acid (18:3)	45.75	7.04	0.30	2.97	4.84
Eicosapentaenoic acid (20:5 <i>n</i> -3)	0.00	0.00	0.00	0.00	18.59
Docosahexaenoic acid (22:6n-3)	0.00	0.00	0.00	0.00	14.67

¹All fat supplements were manufactured as Ca salts by Virtus Nutrition, LLC (Corcoran, CA). Prequel and Strata are Ca salts of PUFA, whereas EnerGII is based on Ca salts of SFA + MUFA.

Diagnostics, Inc., Los Angeles, CA) concentrations. The intra- and interassay CV for haptoglobin were 2.9 and 6.1%, respectively. Plasma cortisol was analyzed within a single assay, and the intra-assay CV was 5.7%.

Statistical Analysis

All cow and calf variables were analyzed with group as the experimental unit and group(treatment × pasture), cow(group), and pasture as random variables. Quantitative data were analyzed using the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC), and binary data were analyzed using the GLIMMIX procedure of SAS. All data was analyzed using gestation days receiving treatment as an independent covariate and Satterthwaite approximation to determine the denominator degrees of freedom for tests of fixed effects. Model statements for cow-related responses included the effects of treatment. Analysis of cow plasma fatty acid profile at calving also included results from d -7 as an independent covariate. Model statements for calfrelated responses analysis included the effects of treatment, calf gender as independent covariate, as well as day and treatment × day interaction for plasma haptoglobin and cortisol analyses Finishing lot and carcass variables analyses also included DOF as an independent covariate. The specified term used in the repeated statement for plasma haptoglobin and cortisol was day, the subject was cow(group), and the covariance structure used was autoregressive, which provided the best fit for these analyses according to the lowest Akaike

information criterion. Results are reported as covariately adjusted least squares means and separated using LSD. Significance was set at $P \le 0.05$, and tendencies were determined if P > 0.05 and < 0.10.

RESULTS AND DISCUSSION

Nutrient composition and profile of diets offered to PUFA- and CON-supplemented cows are described in Tables 1 and 3. Both diets were formulated to represent a typical forage-based diet with limited fat content and provided adequate amounts of energy and CP based on the requirements of pregnant cows during last trimester of gestation (NRC, 2000). It is important to note that both diets included the same amount Ca salts but differed in fatty acid profile. The PUFA treatment was designed to provide equivalent amounts of supplemental *n*-3 and *n*-6, given that both have been shown to impact fetal development (Greenberg et al., 2008; Tanghe and De Smet, 2013). The CON treatment was included to serve as an isolipidic, isocaloric, and isonitrogenous control treatment to PUFA. Hence, results from this experiment should not be associated with differences in total fatty acid intake but with potential fetal programming effects of supplemental *n*-3 and *n*-6 PUFA.

Cow Parameters

Cow age at the beginning of the experiment was similar (P = 0.55) among PUFA- and CON-supplemented cows (Table 4). However, days receiving treatments

 $^{^2}$ Values were obtained from a commercial laboratory wet chemistry analysis (Dairy One Forage Laboratory, Ithaca, NY). The TDN concentration was calculated according to the equations described by Weiss et al. (1992). The NEm concentration was calculated with the following equation (NRC, 2000): NEm = 1.37 ME - 0.138 ME2 + 0.0105 ME3 - 1.12, given that ME = DE \times 0.82 and 1 kg of TDN = 4.4 Mcal DE.

³As percent of total fatty acids. All feedstuffs were analyzed for fatty acid profile according to the procedures described by Moriel et al. (2015).

Table 4. Performance of beef cows receiving diets supplemented with Ca salts of SFA + MUFA (CON) based on palmitic and oleic acids (n = 12), or Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids (n = 12) during the last trimester of gestation^{1,2}

Item	CON	PUFA	SEM	P-value
Cow age, yr	7.6	7.3	0.4	0.55
Days receiving treatments, d	87.0	88.8	0.6	0.02
BW, kg				
Initial (d −7)	584	589	15	0.75
Calving	616	614	11	0.90
BW change	31	25	4	0.20
Weaning (d 280)	575	567	10	0.63
BW change	-41	-45	12	0.43
BCS				
Initial (d −7)	5.01	5.02	0.05	0.89
Calving	5.41	5.46	0.06	0.59
BCS change	0.41	0.42	0.07	0.88
Weaning (d 280)	5.08	5.00	0.08	0.38
BCS change	-0.33	-0.47	0.10	0.26

¹CON = cows received (DM basis) 190 g/cow daily of Ca salts based on palmitic and oleic acids (EnerGII; Virtus Nutrition, LLC, Corcoran, CA); PUFA = cows received (DM basis) 190 g/cow daily of Ca salts based on eicosapentaenoic, docosahexaenoic, and linoleic acids (95 g Prequel + 95 g of Strata; Virtus Nutrition, LLC, Corcoran, CA). Treatments were provided from d 0 (d 195 of gestation) until calving.

²BW and BCS (Wagner et. al., 1988) were recorded prior to the beginning of the experiment (initial; d -7), within 12 h of after calving, and at weaning (d 280).

was greater (P = 0.02) for PUFA-supplemented cows, which was unexpected because all cows conceived during the same timed-AI protocol. Nevertheless, all variables reported herein were covariately adjusted for days receiving treatments to account for a potential biological relevance of this latter outcome. As designed, initial cow BW and BCS (d-7) were similar (P ≥ 0.75) among treatments (Table 4). No treatment effects were detected ($P \ge 0.20$) for any of the subsequent BW and BCS parameters evaluated (Table 4). These outcomes were expected given that PUFA- and CONsupplemented cows consumed similar amounts of energy and CP during late gestation and were managed as a single group from calving until weaning. Others also reported similar BW and BCS at calving between Holstein cows receiving supplements based on PUFA or other fatty acid sources during the prepartum period (Garcia et al., 2014b; Salehi et al., 2016).

Cows assigned to the PUFA and CON supplements had similar ($P \ge 0.11$; Table 5) proportions (as % of total plasma fatty acid) of all plasma fatty acid on d -7, indicating similar fatty acid profiles before treatment administration. At calving, PUFA-supplemented cows had greater (P < 0.01) proportion of plasma vaccenic, linoleic, linolenic, arachidonic, docosapentaenoic, and docosahexaenoic acids as well as total PUFA, n-3, and

n-6 compared with CON-supplemented cows (Table 6). Cows supplemented CON had a greater (P < 0.01) proportion of plasma palmitic, stearic, oleic, eicosapentaenoic, and lignoceric acids as well as total SFA and MUFA compared with PUFA-supplemented cows at calving (Table 6). Overall, these outcomes are in accordance with the fatty acid content and intake of treatments, given that plasma fatty acid profile reflects intake and intestinal fatty acid flow (Klusmeyer and Clark, 1991; Lake et al., 2007; Hess et al., 2008). Furthermore, results from this experiment support those reported by Cook et al. (2017), corroborating that Ca salts of fatty acids can be offered to cows 3 times/wk and still effectively modulate plasma fatty acid profile. The decreased proportion of plasma eicosapentaenoic acid in PUFAsupplemented cows vs. CON-supplemented cows, despite the greater content of this fatty acid in the PUFAbased supplement, was unexpected but can be attributed to its conversion into docosapentaenoic acid (Cook and McMaster, 2002). Accordingly, plasma proportion of docosapentaenoic acid was greater (P < 0.01; Table 6) in PUFA-supplemented cows than in CON-supplemented cows, whereas this fatty acid was not detected in any of the feed ingredients used herein (Table 3).

Calf Birth and Weaning Parameters

No treatment effects were detected $(P \ge 0.16;$ Table 7) for calving rate, proportion of male calves born, and calf birth BW (adjusted or not; BIF, 2010). Therefore, supplementing PUFA or CON to late-gestating cows did not impact calf birth BW, despite treatment differences detected for cow plasma fatty acid profile at calving. At weaning, calves from PUFA-supplemented cows were younger (P = 0.03) compared with cohorts from CON-supplemented cows (Table 7). This 2-d difference in weaning age resulted from the 2-d interval in average calving date between PUFA-supplemented cows and CON-supplemented cows (Table 4) but should be considered biologically irrelevant because it did not impact ($P \ge 0.82$) calf weaning BW (205-d adjusted or not; BIF, 2010). No calf mortality was observed from birth to weaning; hence, weaning rate and proportion of male calves weaned were similar between treatments $(P \ge 0.16)$; data not shown), whereas the latter is known to directly affect weaning BW (Koger and Knox, 1945). Others have also reported similar birth and weaning BW in calves from cows supplemented or not with PUFA during gestation (Hess et al., 2002; Banta et al., 2006; Banta et al., 2011). Collectively, calving and weaning results indicate that supplementing late-gestating beef cows with PUFA did not impact offspring birth BW as well as growth from birth to weaning compared with CON-supplemented cohorts.

Table 5. Initial plasma fatty acid profile (g/100 g of plasma fatty acids) of beef cows assigned to receive diets supplemented with Ca salts of SFA + MUFA (CON) based on palmitic and oleic acids (n = 12), or Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids (n = 12) during the last trimester of gestation^{1,2,3}

Item	CON	PUFA	SEM	P-value
Mystiric acid (14:0)	1.23	1.22	0.04	0.83
Palmitic acid (16:0)	18.09	18.51	0.67	0.59
Palmitoleic acid (16:1)	0.888	0.842	0.055	0.57
Stearic acid (18:0)	34.01	34.17	1.51	0.93
Oleic acid (18:1)	11.88	11.93	0.53	0.92
Vaccenic acid (18:1 trans-11)	0.908	0.903	0.260	0.89
Linoleic acid (18:2 n-6)	11.32	11.54	1.33	0.91
Gamma-linolenic acid (18:3 n-6)	0.271	0.267	0.058	0.96
Linolenic acid (18:3 n-3)	1.98	1.98	0.39	0.99
Arachidic acid (20:0)	0.304	0.296	0.019	0.74
CLA (18:2 <i>n</i> -6 isomers)	0.098	0.098	0.028	0.99
Arachidonic acid (20:4 n-6)	0.502	0.519	0.113	0.91
Eicosapentaenoic acid (20:5 n-3)	0.125	0.078	0.019	0.11
Behenic acid (22:0)	0.760	0.800	0.139	0.84
Adrenic acid (22:4 n-6)	0.077	0.070	0.004	0.21
Docosapentaenoic acid (22:5 n-3)	0.093	0.094	0.029	0.97
Osbond acid (22:5 n-6)	0.026	0.030	0.006	0.68
Docosahexaenoic acid (22:6 n-3)	0.013	0.015	0.007	0.82
Lignoceric acid (24:0)	0.228	0.217	0.016	0.62
Nonidentified fatty acids	1.57	1.25	0.19	0.01
Total SFA	64.49	64.84	2.42	0.90
Total MUFA	19.37	19.14	0.54	0.60
Total PUFA	14.55	14.75	1.95	0.94
Total ω-3	2.25	2.22	0.43	0.96
Total ω-6	12.29	12.53	1.52	0.91

¹CON = cows received (DM basis) 190 g/cow daily of Ca salts based on palmitic and oleic acids (EnerGII; Virtus Nutrition, LLC, Corcoran, CA); PUFA = cows received (DM basis) 190 g/cow daily of Ca salts based on eicosapentaenoic, docosahexaenoic, and linoleic acids (95 g Prequel + 95 g of Strata; Virtus Nutrition, LLC, Corcoran, CA). Treatments were provided from d 0 (d 195 of gestation) until calving.

²Blood samples were collected from all cows on d -7 of the experiment and analyzed for fatty acid profile according to the procedures described by Garcia et al. (2014a)

³SFA = mystiric, palmitic, stearic, arachidic, behenic, and lignoceric acids; MUFA = palmitoleic, oleic, and vaccenic acids; PUFA = linoleic, gamma-linolenic, linolenic, CLA, arachidonic, eicosapentaenoic, adrenic, docosapentaenoic, osbond, and docosahexaenoic acids.

Calf Preconditioning Parameters

Maternal nutrition during gestation has been shown to impact steroidogenesis in the offspring, including plasma cortisol response to weaning (Long et al., 2010; Marques et al., 2016a). However, no treatment effects were detected (P = 0.20) herein for plasma cortisol (Table 7), which increased (day effect; P < 0.01) for both treatments on weaning (28.5, 31.7, 32.7, and 28.4 ng/mL on d 280, 282, 285, and 288, respectively; SEM = 1.2). A treatment × day interaction was

Table 6. Plasma fatty acid profile (g/100 g of plasma fatty acids) at calving of beef cows receiving diets supplemented with Ca salts of SFA + MUFA (CON) based on palmitic and oleic acids (n = 12), or Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids (n = 12) during the last trimester of gestation 1,2,3

Item	CON	PUFA	SEM	P-value
Mystiric acid (14:0)	0.893	0.920	0.040	0.54
Palmitic acid (16:0)	26.74	17.92	1.68	< 0.01
Palmitoleic acid (16:1)	0.886	0.951	0.095	0.33
Stearic acid (18:0)	25.87	18.76	2.38	< 0.01
Oleic acid (18:1)	13.55	6.99	0.23	< 0.01
Vaccenic acid (18:1 trans-11)	0.553	0.794	0.022	< 0.01
Linoleic acid (18:2 n-6)	19.52	38.74	3.14	< 0.01
Gamma-linolenic acid (18:3 <i>n</i> -6)	0.198	0.116	0.080	0.15
Linolenic acid (18:3 n-3)	2.014	3.733	0.595	< 0.01
Arachidic acid (20:0)	0.115	0.120	0.010	0.73
CLA (18:2 <i>n</i> -6 isomers)	0.082	0.112	0.025	0.14
Arachidonic acid (20:4 n-6)	0.548	2.079	0.189	< 0.01
Eicosapentaenoic acid (20:5 n-3)	0.099	0.013	0.026	< 0.01
Behenic acid (22:0)	0.604	0.396	0.175	0.10
Adrenic acid (22:4 n-6)	0.020	0.014	0.003	0.18
Docosapentaenoic acid (22:5 n-3)	0.101	0.443	0.064	< 0.01
Osbond acid (22:5 n-6)	0.026	0.029	0.005	0.68
Docosahexaenoic acid (22:6 n-3)	0.004	0.573	0.047	< 0.01
Lignoceric acid (24:0)	0.070	0.039	0.007	< 0.01
Nonidentified fatty acids	0.795	0.670	0.077	0.30
Total SFA	58.84	41.65	4.17	< 0.01
Total MUFA	17.82	11.86	0.28	< 0.01
Total PUFA	22.65	44.89	4.06	< 0.01
Total ω -3	2.25	4.80	0.65	< 0.01
Total ω-6	20.40	41.09	3.42	< 0.01

¹CON = cows received (DM basis) 190 g/cow daily of Ca salts based on palmitic and oleic acids (EnerGII; Virtus Nutrition, LLC, Corcoran, CA); PUFA = cows received (DM basis) 190 g/cow daily of Ca salts based on eicosapentaenoic, docosahexaenoic, and linoleic acids (95 g Prequel + 95 g of Strata; Virtus Nutrition, LLC, Corcoran, CA). Treatments were provided from d 0 (d 195 of gestation) until calving.

²Blood samples were collected from all cows (n = 48 per treatment) within 12 h after calving. Values reported herein were covariately-adjusted to values obtained on d -7. All samples were analyzed for fatty acid profile according to the procedures described by Garcia et al. (2014a)

³SFA = mystiric, palmitic, stearic, arachidic, behenic, and lignoceric acids; MUFA = palmitoleic, oleic, and vaccenic acids; PUFA = linoleic, gamma-linolenic, linolenic, CLA, arachidonic, eicosapentaenoic, adrenic, docosapentaenoic, osbond, and docosahexaenoic acids.

detected (P = 0.05) for plasma haptoglobin concentrations (Fig. 1), which also increased for both treatments on weaning (day effect; P < 0.01) but was greater (P = 0.05) in calves from CON-supplemented cows on d 282. The day effects reported herein for plasma cortisol and haptoglobin concentrations were expected, based on the neuroendocrine stress response and acutephase protein reaction elicited by weaning and vaccination against BRD pathogens (Arthington et al., 2013; Rodrigues et al., 2015). However, the acute-phase

Table 7. Calving, weaning, and preconditioning outcomes from beef cows receiving diets supplemented with Ca salts of SFA + MUFA (CON) based on palmitic and oleic acids (n = 12), or Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids (n = 12) during the last trimester of gestation 1

Item	CON	PUFA	SEM	P-value
Calving results				
Calving rate, %	100	95.7	2.6	0.16
Percent of male calves born	46.8	56.8	7.5	0.34
Calf birth BW, kg	40.9	41.7	0.6	0.44
Adjusted calf birth BW,2 kg	41.3	42.0	0.6	0.42
Weaning results				
Percent of male calves weaned	46.8	56.8	7.5	0.34
Calf weaning age, d	193	191	1	0.03
Calf weaning BW, kg	241	242	3	0.82
Calf 205-d adjusted weaning BW,2 kg	258	259	3	0.86
Preconditioning results				
Plasma cortisol, ng/mL	32.3	30.3	1.2	0.20
Treated for BRD signs, ³ %	6.8	3.8	3.8	0.55
Preconditioning ADG, kg/d	0.43	0.50	0.05	0.31
End of preconditioning BW,4 kg	261	265	3	0.29

¹CON = cows received (DM basis) 190 g/cow daily of Ca salts based on palmitic and oleic acids (EnerGII; Virtus Nutrition, LLC, Corcoran, CA); PUFA = cows received (DM basis) 190 g/cow daily of Ca salts based on eicosapentaenoic, docosahexaenoic, and linoleic acids (95 g Prequel + 95 g of Strata; Virtus Nutrition, LLC, Corcoran, CA). Treatments were provided from d 0 (d 195 of gestation) until calving.

protein response is also stimulated by elevated circulating cortisol (Cooke and Bohnert, 2011; Cooke et al., 2012), whereas treatment differences for plasma haptoglobin concentration on d 282 were not accompanied by a similar cortisol response. These outcomes indicate that PUFA supplementation to late-gestating cows did not impact the steroidogenesis required to cope with the stress of weaning procedures in the offspring but altered the resulting plasma haptoglobin protein response (Carroll and Forsberg, 2007). The reason for this latter outcome is unknown but could be associated with immunomodulatory effects of PUFA as previously reported by our group (Araujo et al., 2010; Cooke et al., 2011). Nevertheless, research is warranted to determine potential mechanisms by which PUFA supplementation to late-gestating beef cows impacts the stress-induced haptoglobin response in the offspring.

During the 45-d preconditioning period, no treatment effects were detected ($P \ge 0.29$) for incidence of calves that required treatment for BRD, ADG, and BW at the end of preconditioning period (Table 7). Furthermore, all calves diagnosed with BRD required only 1 treat-

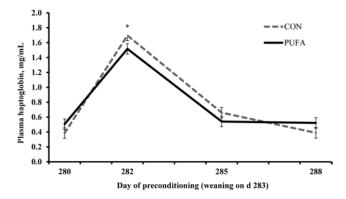


Figure 1. Plasma haptoglobin concentration from weaned calves (d 280 of the experiment) from beef cows receiving diets supplemented with 190 g/cow daily of Ca salts of SFA + MUFA based on palmitic and oleic acids (CON; n = 12) or 190 g/cow daily of Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids (n = 12) during the last trimester of gestation (from d 195 of gestation until calving). A treatment × day interaction was detected (P < 0.05). *P = 0.05.

ment with antimicrobial to eliminate BRD signs. These results indicate that calf preconditioning responses were not impacted by treatments despite differences detected for plasma haptoglobin concentration (Fig. 1), which has been negatively associated with performance and immune parameters in weaned cattle (Berry et al., 2004; Qiu et al., 2007; Araujo et al., 2010). Perhaps the magnitude of haptoglobin differences between calves from CON-supplemented cows and calves from PUFA-supplemented cows was not sufficient to impact calf growth and health responses during preconditioning.

Calf Feedlot and Carcass Parameters

During the growing phase, no treatment effects were detected ($P \ge 0.52$) for initial BW and proportion of calves treated for BRD signs, whereas none of the calves diagnosed with BRD required a second antimicrobial treatment. Calves from PUFA-supplemented cows had greater (P = 0.05) ADG and tended to be heavier (P = 0.09) at the end of the growing phase compared with calves from CON-supplemented cows (Table 8).

During the finishing phase, DOF and the proportion of animals treated for BRD signs were similar ($P \ge 0.16$) among treatments (Table 8), and no second antimicrobial administration was required in calves diagnosed with BRD. Calves from PUFA-supplemented cows tended to have greater (P = 0.06) ADG and were heavier (P = 0.05) at the end of the finishing phase compared with calves from CON-supplemented cows (Table 8). Percentage of total or male calves slaughtered did not differ among treatments ($P \ge 0.16$; data not show) and were equal (P = 1.00) to calving rate and percent of male calves born (Table 7), respectively, given that zero calf mortality was observed during the experimental period. Upon slaughter, HCW and marbling were greater ($P \le 0.16$)

²Calculated according to BIF (2010).

³Calves were classified as positive for BRD signs according to the subjective criteria described by Berry et al. (2004), and received 1 mL/10 kg of BW of Hexasol LA Solution (Norbrook Inc. USA; Overland Park, KS 6).

⁴Collected upon growing lot (Top Cut; Echo, OR) arrival.

Table 8. Feedlot performance and carcass characteristics of feeder cattle from beef cows receiving diets supplemented with Ca salts of SFA + MUFA (CON) based on palmitic and oleic acids (n = 12), or Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids (n = 12) during the last trimester of gestation¹

Item	CON	PUFA	SEM	P-value
Growing phase performance				
Initial growing BW, kg	248	250	3	0.68
Treated for BRD signs, ² %	38.3	31.8	7.1	0.52
Growing phase ADG, kg/d	1.12	1.22	0.03	0.05
BW at the end of growing phase, kg	383	397	6	0.09
Finishing phase performance				
Days on feed, d	127	126	1	0.34
Treated for BRD signs, ² %	2.2	2.2	2.2	0.94
BW at the end of finishing phase, ³ kg	621	646	9	0.05
Finishing phase ADG, kg/d	1.87	1.98	0.04	0.06
Carcass characteristics ⁴				
HCW, kg	391	407	6	0.05
Back fat, cm	1.74	1.82	0.09	0.38
LM area, cm ²	89.6	92.3	1.2	0.10
КРН, %	2.15	2.13	0.07	0.85
Marbling	489	539	16	< 0.01
Yield grade	3.50	3.56	0.11	0.63
Retail product, %	48.6	48.4	0.3	0.56
Choice, %	93.5	100.0	2.7	0.09

¹CON = cows received (DM basis) 190 g/cow daily of Ca salts based on palmitic and oleic acids (EnerGII; Virtus Nutrition, LLC, Corcoran, CA); PUFA = cows received (DM basis) 190 g/cow daily of Ca salts based on eicosapentaenoic, docosahexaenoic, and linoleic acids (95 g Prequel + 95 g of Strata; Virtus Nutrition, LLC, Corcoran, CA). Treatments were provided from d 0 of the experiment (d 195 of gestation) until calving. Calves were assigned to the growing phase from d 325 to 424 of the experiment, and finishing phase from d 425 until slaughter.

²Calves were classified as positive for BRD symptoms according to the DART system (Zoetis, Florham Park, NJ), and received medication according to the feedyard management criteria.

³Calculated based on HCW (assuming 63% dressing; Loza et al., 2010).

⁴Backfat thickness measured at the 12th rib; marbling score: 400 =

Small00, 500 = Modest00; 600 = Medium00; yield grade calculated as reported by Lawrence et al. (2010); USDA retail yield equation = $51.34 - (5.78 \times \text{backfat}) - (0.0093 \times \text{HCW}) - (0.462 \times \text{KPH}) + (0.74 \times \text{LM} \text{ area})$.

0.05) and LM area tended to be greater (P=0.10) in calves from PUFA-supplemented cows than in calves from CON-supplemented cows (Table 8). Calves from PUFA-supplemented cows also tended (P=0.09) to have greater percent of carcasses graded as Choice (Table 8), although the vast majority of calves from CON-supplemented cows also graded Choice. No treatment differences were detected ($P \ge 0.38$) for the remaining carcass merit traits evaluated (Table 8). To our knowledge, these results are novel and indicative of fetal programming effects from supplementing PUFA to lategestating beef cows (Funston et al., 2010).

Maternal nutrition impacts fetal skeletal muscle development through hyperplasia and hypertrophy, result-

ing in permanent effects on postnatal growth and performance (Zhu et al., 2004; Nathanielsz et al., 2007; Du et al., 2010). During late gestation, however, only muscle hypertrophy and adipocyte development are significantly influenced in the fetus by maternal nutritional status, with direct consequences on lifelong growth and intramuscular fat deposition (Harper and Pethick, 2004; Du et al., 2010, 2011). Corroborating the treatment differences reported herein for ADG, HCW, LM area, and carcass marbling, PUFA have been shown to impact muscle and adipocyte function in developing tissues. Hiller et al. (2012) reported that *n*-3 fatty acid positively regulates the expression of genes associated with muscle development and function but reduced expression of genes regulating lipogenesis and fatty acid accumulation in the LM to favor metabolism of muscle cells. On the other hand, n-6 fatty acid has been shown to have adipogenic effects by increasing the expression of PPARy in muscle tissues, a key promoter of adipocyte differentiation and marbling in cattle (Moriel et al., 2014). Mangrum et al. (2016) reported that supplementing n-6 fatty acid to young cattle enhanced intramuscular adipocyte development, resulting in increased carcass marbling on slaughter. Alternatively, treatment effects detected on growth and carcass traits also may have been elicited by greater DMI during the growing and finishing phases in calves from PUFA-supplemented cows (NRC, 2000), given that maternal dietary fat content appears to impact appetite regulation in the offspring.(Gupta et al., 2009). Hence, the improvement in feedlot growth and carcass quality in calves from PUFA-supplemented cows should be attributed to the combination of supplemental n-3 and n-6, whereas the specific role of each PUFA deserves further investigation. By providing these PUFA during late gestation, it can be speculated that accumulation of these fatty acids into fetal tissues was increased, enhancing development of muscle and adipose cells and modulating DMI regulation, which translated into increased carcass growth and marbling when offspring were provided highenergy anabolic feedlot diets (Harper and Pethick, 2004).

Overall Conclusions

Supplementing forage-fed beef cows during late gestation with Ca salts of PUFA based on equivalent amounts of *n*-3 and *n*-6 fatty acids did not impact cow performance during gestation, calving rate, or calf birth BW. At calving, the proportion of plasma *n*-3 and *n*-6 fatty acids was greater in PUFA-supplemented cows than in CON-supplemented cows. No major differences were observed in offspring performance, health, and immune parameters from birth to weaning and subsequent 45-d preconditioning. However, after being exposed to a high-energy feedlot diet, HCW was 16 kg heavier and

carcass marbling increased from small to modest when comparing calves from PUFA-supplemented cows with calves from CON-supplemented cows. These results are indicative of programming effects on postnatal offspring growth and health resulting from PUFA supplementation to late-gestating cows (Funston et al., 2010), although the mechanism underlying these effects, including the specific role of *n*-3 and *n*-6 fatty acids, also warrants investigation. Nevertheless, these outcomes are novel and indicate that supplementing gestating beef cows with Ca salts of PUFA based on eicosapentaenoic, docosahexaenoic, and linoleic acids might be a feasible alternative to optimize offspring productivity in beef production systems.

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