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Feeding excess crude protein to gestating and lactating beef heifers: Impact on parturition, milk composition, ovarian function, reproductive efficiency and pre-weaning progeny growth

P.J. Gunn [a,1], J.P. Schoonmaker [a], R.P. Lemenager [a], G.A. Bridges [b,*]

[a] Department of Animal Sciences, Purdue University, 915W. State St., West Lafayette, IN 47907, USA
[b] North Central Research and Outreach Center, University of Minnesota, 1861E. Highway 169, Grand Rapids, MN 55744, USA

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ABSTRACT

Angus–Simmental beef heifers confirmed pregnant to a single sire ($n=78$) were used to determine the effects of feeding excess CP during late gestation and early lactation on parturition, milk composition, reproduction, and pre-weaning progeny growth. At 192 d in gestation, heifers were allotted by BW within BCS to receive either a corn silage-based control diet balanced to meet or exceed NRC recommendations (2000) (CON), or a diet that considerably exceeded CP requirements consisting of corn stover and DDGS, where DDGS were fed as a primary energy source (HP). Diets were formulated to provide similar daily energy intake (2.09 Mcal/d NE_g). Dietary treatments were stopped at 118 ± 4 d postpartum (DPP) and cow/calf pairs were comingled. Calving parameters and pre-weaning calf performance were assessed. At 64 ± 4 DPP, milk samples were collected to determine composition and fatty acid profile. At 32 ± 4 DPP, a single follicular wave was mapped via ultrasonography in 12 anestrous heifers per treatment. Starting 34 ± 4 DPP, blood samples were taken every 10 d until synchronization to determine resumption of estrous cyclicity. Gestation was longer ($P=0.02$) and incidence of dystocia was greater ($P=0.003$) for HP than CON, respectively. Birth BW was greater ($P<0.001$) in HP than CON progeny. However, milk fat, total solids and energy corrected milk production were greater ($P\leq 0.03$) in CON than HP, respectively. In contrast, milk urea N, MUFA, PUFA, and CLA content was greater ($P<0.01$) in HP than CON, respectively. During the anestrous period, dominant and secondary follicle diameter was greater ($P\leq 0.05$) and follicle wavelength tended ($P=0.08$) to be greater in HP than CON, respectively. Proportion of cyclic heifers at estrous synchronization did not differ between treatments, but HP tended ($P=0.10$) to resume estrous activity at fewer DPP than CON. Timed-AI and breeding season pregnancy rates did not differ ($P\geq 0.30$) between the HP and CON treatment. Calf BW was greater throughout the pre-weaning period ($P<0.001$) for HP than CON progeny. Results indicate that feeding DDGS as a primary energy source to first-parity heifers during late gestation and early lactation altered ovarian function and resulted in increased pre-weaning growth performance of progeny.

* Corresponding author. Tel.: +1 218 327 4615.
E-mail address: gbridges@umn.edu (G.A. Bridges).
[1] Current address: Department of Animal Science, Iowa State University, Ames, IA 50011, USA.

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1. Introduction

Until recent years, overfeeding CP to beef cattle was not typically of concern or a common practice due to cost. However, with the increased availability of cost-effective byproduct feeds that are rich in both energy and protein, the potential exists to use these feedstuffs as a primary dietary energy source. Thus, diets rich in byproducts may grossly exceed the CP requirements of the animal and could change the balance between protein and energy, alter normal energy partitioning, and affect both performance and reproduction. Dried distillers grains with solubles (DDGS) is a byproduct feed that is readily available in many areas, and volatile commodity prices of traditional dietary energy sources such as corn have increased the utilization of DDGS in beef cattle diets. However, feeding increased amounts of DDGS not only leads to excessive dietary CP concentrations, but also increases dietary fat from corn oil, which is rich in polyunsaturated fatty acids and may also affect performance and reproductive parameters.

Excess dietary CP has been correlated with decreased conception rates in dairy cattle (Butler et al., 1996), which may be a result of a shift in uterine pH (Rhoads et al., 2004) and altered uterine function (Elrod and Butler, 1993), or impaired embryo viability (Rhoads et al., 2006). However, when the dietary CP source also provides additional dietary fat, it is unclear if the beneficial effects of increased unsaturated fatty acids could potentially outweigh the negative effects of excessive CP. Previous studies have indicated that increased concentrations of unsaturated fatty acids in the diet may increase ovulatory follicle growth in dairy cattle (Bilby et al., 2006) and improve pregnancy rates in *Bos indicus* (Lopes et al., 2009) and dairy (Reis et al., 2012) cattle. Yet, research on feeding a feedstuff rich in CP and unsaturated fatty acids, such as DDGS, as an energy source to lactating beef cows has been inadequate to date. To our knowledge, feeding DDGS as a primary energy source to first parity heifers, thus exceeding protein requirements (by a minimum of 150%), through late gestation and early lactation has not been investigated. Therefore, the objectives of this study were to assess the effects of feeding excessive CP via supplementation with DDGS during the last trimester of pregnancy and early lactation on parturition, milk, and reproductive parameters of the dam. Moreover, because altered maternal diet during pregnancy and the neonatal period can also affect subsequent progeny post-natal growth through developmental programming (Reynolds et al., 2010), the effects of a maternal diet rich in CP and fat on pre-weaning progeny growth was also studied. We hypothesized that a diet which provided excess CP in combination with increased fat would alter birth BW and milk composition, improve follicular growth, and have no effect on resumption of estrous cyclicity, but suppress TAI pregnancy rates.

2. Materials and methods

2.1. Animals and diets

All cows were handled in accordance with procedures approved by the Purdue Animal Care and Use Committee and conducted at the Purdue Animal Sciences Research and Education Center in West Lafayette, Indiana. Yearling, Angus–Simmental heifers of similar genetic background [$n=78$; BW$=518\pm6$ kg; BCS$=5.1\pm0.03$ (1=emaciated, 9=obese; Wagner et al., 1988) at 192 d of gestation] were used in a randomized complete block design study to elucidate the effects of feeding excess CP, during late gestation and early lactation on heifer body weight and composition changes, calving and lactation parameters, and pre-weaning progeny performance. Heifers were used as they are a more nutritionally sensitive model and use of these females allowed us to reduce variation in age, weight and BCS at trial initiation.

Fig. 1 illustrates the timeline of treatment implementation and evaluations conducted. As virgin heifers, these females were timed artificial inseminated (TAI) on the same day to a single sire. Only heifers confirmed pregnant to TAI were utilized for the study. At 192 d of gestation, heifers were blocked by BCS and stratified by BW within BCS and assigned to 1 of 2 diets where they were pen fed in a singular group for each treatment: (1) a corn silage-based TMR Control (CON), or (2) ad libitum access to corn stover bales with DDGS supplemented to meet the energy requirements of a heifer in her third trimester of gestation and lactation but exceed requirements for CP (HP; Table 1). Diets were formulated to deliver a similar amount of NE_g per day and to meet or exceed the heifer protein, vitamin and mineral requirements during late gestation, and subsequently early lactation (NRC, 2000). All diets were formulated using individual ingredient chemical composition analysis obtained by wet chemistry methods (AOAC, 1990) before the start of the trial (Sure-Tech Laboratories, Indianapolis, IN).

Diets were formulated to deliver similar amounts of NE_g, thus facilitating a prepartum ADG of 0.45 kg, and

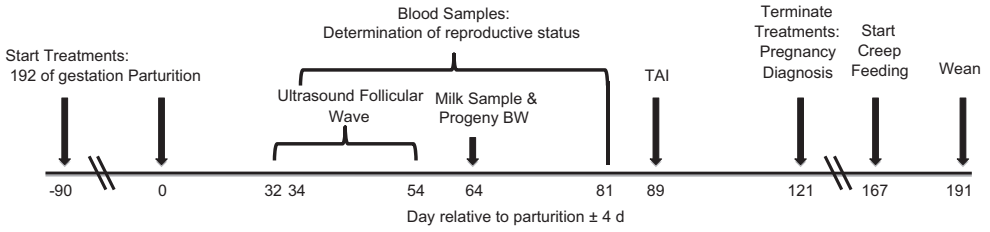

Fig. 1. Timeline of treatment implementation and evaluations conducted. Treatments were initiated at 192 d of gestation in all females. Due to differences in date of parturition, all days listed are average days relative to parturition ±4 d.

Table 1
Dietary ingredients, nutrient composition and calculated nutrient intake of diets fed to yearling beef heifers from 192 days in gestation through 118 ± 4 days in lactation.

Item	Treatment[a]			
	Prepartum		Postpartum	
	CON	HP	CON	HP
Ingredient, formulated (kg DM/d)				
Dried distiller's grains[b]	–	5.13	–	6.00
Cracked corn	–	–	1.00	–
Corn silage	9.09	–	5.42	–
Grass haylage	–	–	6.09	–
Corn stover[c]	–	6.34	–	5.96
Limestone	–	0.22	–	0.16
Urea	0.09	–	–	–
Vitamin and mineral supplement	0.11	0.11	0.11	0.11
Formulated nutrient intake[d,e]				
CP (g/d)	1021	1783	1409	2021
NE_g (Mcal/d)	2.06	2.11	2.04	1.97
Fat (g/d)	280	631	433	720
Ca (g/d)	40	102	92	99
P (g/d)	28	63	40	62
S (g/d)	15	58	26	57
Estimated DMI (kg/d)[d,f]				
Dried distiller's grains	–	5.10	–	6.0
Cracked corn	–	–	0.96	–
Corn silage	8.99	–	5.31	–
Grass haylage	–	–	5.95	–
Corn stover	–	6.5	–	6.8
Limestone	–	0.22	–	0.16
Urea	0.09	–	–	–
Mineral supplement	0.11	0.11	0.11	0.11
Calculated nutrient intake[g]				
CP (g/d)	1014	1781	1379	2057
RUP (g/d)[h]	305	1017	372	1224
RDP (g/d)[i]	709	764	1007	833
NE_g (Mcal/d)	1.96	2.13	1.98	2.11

[a] CON=control; HP=dried distiller's grains supplemented as an energy source.
[b] Dried distiller's grain with solubles. Fatty acid composition (g/100 g): C16:0=12.3%, C18:0=1.7%, C18:1=25.1%, C18:2*n*–6=57.8%, C18:3=1.5%, and other=1.6%.
[c] Corn stover analysis (DM basis): 4.3% CP, 0.38% Ca, 0.10% P, and 0.55 Mcal/kg NE_g.
[d] Expressed on DM basis.
[e] Calculated dietary chemical composition based on analysis of individual ingredients.
[f] DMI was measured for CON total mixed ration delivered and HP supplement delivered. Corn stover DMI was estimated based on weight of bales delivered and uncorrected for wastage.
[g] Calculated daily nutrient intake based on estimated DMI.
[h] Calculated intake of rumen undegradable protein.
[i] Calculated intake of rumen degradable protein.

allowing heifers to achieve 85% of mature BW at parturition. Subsequent postpartum ADG was targeted at 0.39 kg. For the HP treatment, prepartum daily corn stover DMI was estimated using the following formula (NRC, 2000) for pregnant beef cows in the last two-thirds of pregnancy:

$$\mathrm{DMI} = \left(\frac{\mathrm{SBW}^{0.75} \times (0.04997\ \mathrm{NE_m} + 0.04631)}{\mathrm{NE_m}}\right) \times (\mathrm{TEMP1}) \times (\mathrm{MUD1}),$$

where SBW was shrunken BW, NE_m was the net energy for maintenance of the corn stover, TEMP1 (1.05) and MUD1 (0.85) were adjustments for temperature and mud, respectively.

For the postpartum HP diet, daily corn stover DMI was estimated using the following formula (NRC, 2000) for non-pregnant beef cows:

$$\mathrm{DMI} = \left(\frac{(\mathrm{SBW}^{0.75} \times 0.04997 \times \mathrm{NE_m^2} + 0.03840}{\mathrm{NE_m}}\right) \times (\mathrm{TEMP1}) \times (\mathrm{MUD1}) + (0.2 \times \mathrm{Yn})),$$

where adjustments for TEMP1 and MUD1 were 0.90 and 0.85, respectively; Yn was milk production in kg.

Once corn stover DMI was estimated for HP diets, DDGS were added to meet the energy requirement for desired gains. CON diets and intakes were then formulated to be isocaloric with the HP diet. Corn stover bales were presented for consumption in a combination of ring and rectangle-style bale feeders. Weights of bales were recorded before delivery; and although refusal and waste was not recorded because it could not be accurately quantified,

wastage was minimized by feeding new bales only once previous bales were at least 90% removed from the feeder. Dietary and feed delivery adjustments were made to the CON treatment as needed to maintain similar BW between treatments throughout the study. The CON diet and HP supplement were delivered in concrete fence-line bunks once daily at approximately 0900. Individual feed ingredients were analyzed weekly for DM via forced air oven at 60 °C for 72 h to adjust intake for dietary moisture content.

Dietary treatments finished at 118 ± 4 days postpartum (DPP), which coincided with initial TAI pregnancy diagnosis. Once treatments were terminated, all cow-calf pairs across treatments were commingled, placed on pasture, and managed as a singular group until weaning which occurred at 191 ± 4 d postpartum.

Initial BW was determined by taking the average preprandial BW measured on 2 consecutive days prior to the start of dietary treatments. Initial BCS was taken the day before treatment initiation. Subsequent BW and BCS were assessed monthly throughout the treatment period with BCS assessment conducted by the same person at all time points throughout the study. The final prepartum BW and BCS was taken 14 d before the expected calving due date and assessments resumed 26 d later after all heifers had calved. Final treatment BW and BCS were assessed at 118 ± 4 DPP. In addition, first-parity heifer post-treatment BW and BCS were recorded at 167 and 191 ± 4 DPP to aid in interpretation of progeny growth between the treatment period and weaning. For purposes of data analysis, prepartum heifer BW was adjusted for gravid uterine wt (GUW, Ferrell et al., 1976). One CON and two HP heifers aborted their fetus between final pregnancy diagnosis and beginning of the trial. These heifers were completely removed from the data set. Females that gave birth to a stillborn calf or experienced neonatal mortality (HP $n=1$; CON $n=5$) were removed from the trial, but their prepartum performance data remained in the data set.

2.2. Parturition

Beginning 14 d before the projected calving date and throughout the 18 d calving period, heifers were monitored for signs of parturition every 3 h. Once labor began and either the allantois or amnion was presented, heifers were given 1 h to calve or make significant progress as indicated by presentation of a portion of the fetus. Females that did not calve or make significant progress within 1 h were given assistance. Females that did progress were monitored hourly and given assistance if progress slowed or halted. After parturition, but before nursing, sex of the calf was recorded, BW of the calf was measured, and a calving difficulty score (1=no assistance; 2=easy pull; 3=mechanically assisted pull; 4=abnormal presentation; and 5=caesarian-section) was assigned. Calves were monitored to ensure they nursed and received colostrum within 12 h after birth. A calf vigor score (1=nursed on own immediately; 2=nursed on own, but slow to start; 3=required assistance to nurse; 4=died shortly after birth) was then assigned within 12 h after birth. In addition, cow udder score (1=ideal; 2=not ideal, but calf nursed on own; 3=required intervention, cull cow with no daughters retained; 4=worst case; calf could not latch; cull cow with no daughters retained) was assessed 48 h postpartum, allowing for parturition induced mammary edema to subside. Bull calves were castrated within 48 h of birth. For purposes of data analysis, dystocia was categorized on a yes/no basis. Therefore, any heifer that was given assistance, and thus had a calving difficulty score of greater than 1 was given a "yes" for dystocia. One HP and 2 CON treated females gave birth to stillborn calves. For those heifers, calf sex, BW at birth, and calving ease was recorded. Calf vigor and udder score for these females was not recorded.

2.3. Milk

At 64 ± 4 DPP, preprandial milk samples were collected from a subsample of 26 CON and 31 HP treated females. The left rear quarter of the udder was hand-milked until no more milk could be produced. Collected milk was mixed and two aliquots were transferred to 50-mL polystyrene conical tubes and stored at −20 °C for analysis of fatty acid profile. A third aliquot was shipped that day and analyzed for milk protein, milk fat, milk lactose, total solids, and milk urea nitrogen (MUN) by a third-party laboratory (Dairy One Cooperative, Ithaca, NY).

The following day, milk production was assessed in all lactating heifers on study (CON $n=33$; HP $n=36$) via a 24-h weigh-suckle-weigh procedure (WSW; Lemenager et al., 1978). Briefly, calves were separated from their dams at 0200, allowed back to nurse the cows dry at 0800, and again separated, serving as the empty baseline. At 1400, calves were weighed before nursing, allowed to nurse their dam, and once nursing was completed, immediately reweighed. The difference between post- and preprandial calf BW was the calculated milk production for each 6 h interval. This procedure was repeated over 3 more consecutive 6-h intervals, and a 24-h milk production was calculated by adding the 4 production periods together. Cows were allowed access to diets and water between nursing periods. Calves were not allowed access to any feed or water during the WSW procedure, but were given access to shade. Utilizing total milk production data and milk composition, energy corrected milk (ECM) production was calculated using the following equation (Bernard, 1997):

$$\mathrm{ECM} = (0.3246 \times \mathrm{kg\ \ totalmilk/d}) + (12.86 \times \mathrm{kg\ \ milkfat/d}) + (7.04 \times \mathrm{kg\ \ milkprotein/d}).$$

Milk samples collected on 64 ± 4 DPP were evaluated for fatty acid profile. Total lipid was extracted with a chloroform and methanol (2:1, v-v) mixture and quantified (Folch et al., 1957). Lipids were derivatized to methyl esters according to Lepage and Roy (1986). Fatty acid methyl esters were analyzed by a gas chromatography (GC; model 3900, Varian) equipped with a Supelco SP-2380 column (100 m × 0.25 mm i.d. × 0.20 μm film thickness) and a flame ionization detector. Short chain fatty acids were categorized as those between C4:0 and C8:0, medium chain fatty acids were categorized as those between C10:0 and C15:0, and long chain fatty acids were categorized as C16:0 and greater.

2.4. Ovarian follicular wave characterization

At 32 ± 4 DPP, a subsample of 12 anestrous heifers from each treatment were randomly selected to undergo daily transrectal ultrasonography (variable MHz linear array transducer, MicroMaxx, Sonosite, Bothell, WA) for a period of time sufficient to identify and characterize a single anestrous follicular wave in its entirety. Scans were conducted by a single trained technician. The number, location, and diameter of all ovarian follicles ≥ 3 mm in diameter was recorded by drawing sketches of both ovaries during each examination. The dominant follicle was recognized as the largest growing follicle during the follicular wave, and the secondary follicle was the second largest growing follicle identified during the wave of interest. Maximum diameter of the dominant and secondary follicle was defined as the largest diameter achieved during the follicular wave and was determined by measuring the largest cross-sectional diameter of the follicle with the caliper function of the ultrasound. The diameter at which the dominant follicle attained dominance was defined as the cross-sectional measurement taken on the first day in which the dominant follicle was at least 1 mm larger in diameter than any other growing follicle of that wave. Duration of dominance was defined as the number of days from attainment of dominance until emergence of the subsequent follicular wave. Emergence of the follicular wave of interest and subsequent wave were determined retrospectively and defined as the day on which the dominant follicle could be traced back to be a member of a cohort of growing follicles 4 mm in diameter. Duration of the follicular wave, also referred to as wavelength, was determined to be the number of days from emergence of the follicular wave of interest until emergence of the subsequent wave. The mean number of antral follicles per day over the duration of the wave was also calculated. Daily ultrasound evaluation of individual animals was terminated once the dominant follicle of the subsequent follicular wave could be identified. Three heifers from the HP treatment and 2 heifers from the CON treatment ovulated during follicular wave characterization, and therefore, their follicular data were removed from the data set and not used for statistical analysis.

2.5. Resumption of cyclicity

Starting at 34 ± 4 DPP, blood samples were collected by 10-d intervals until estrous synchronization (81 ± 4 DPP) for determination of approximate duration of the anestrous period. Samples were collected via coccygeal venipuncture in 6 mL tubes containing EDTA (BD Vacutainer™; Becton-Dickinson, Franklin Lakes, NJ) and immediately placed on ice. Samples were centrifuged at 1750 × *g* for 25 min at 4 °C; plasma was recovered, transferred to 5 mL polystyrene tubes and frozen at −20 °C until analyzed for progesterone concentration. Heifers that had progesterone concentrations ≥ 1 ng/mL on any given sampling day were determined to have resumed estrous cycles. If plasma samples had ≥ 1 ng/mL, but < 2 ng/mL of progesterone, the date on which cyclicity resumed was determined to be 5 d before the sampling date. If plasma samples had ≥ 2 ng/mL of progesterone, the date on which cyclicity resumed was determined to be 10 d before the sampling date. The anestrous period for each heifer was calculated as the number of days between parturition and resumption of estrous cycles. Furthermore, all subsequent plasma samples obtained prior to estrous synchronization were analyzed for progesterone to confirm that heifers remained cycling.

Progesterone concentration was determined using a commercially available RIA kit (Coat-A-Count, Siemens Medical Solutions Diagnostics, Los Angeles, CA). Across 2 assays, the average intra-assay CV was 1.7% and the inter-assay CVs for pooled plasma samples containing 0.19 ng/mL and 6.35 ng/mL of progesterone, were 2.3% and 1.7%, respectively. The average sensitivity across assays was 0.16 ng/mL (95% confidence interval).

2.6. Blood urea nitrogen

At −35, 56, and 80 ± 4 days relative to parturition, jugular blood samples were collected for analysis of blood urea nitrogen (BUN). One sample was collected via venipuncture into 6 mL serum tubes (BD Vacutainer™; Becton-Dickinson, Franklin Lakes, NJ) and immediately placed on ice. Samples were given 4 h to clot and were then centrifuged at 1750 × *g* for 25 min at 4 °C; serum was recovered, transferred to 5 mL polystyrene tubes and frozen at −20 °C until analyzed for BUN. Serum samples were analyzed for BUN concentrations using a commercial kit (Urea Nitrogen Procedure No. 0580, Stanbio Laboratory, Boerne, TX). Samples were read in 96-well polystyrene plates (Becton, Dickinson and Co., Franklin Lakes, NJ) on an Opsys MR microplate reader (Dynex Technologies Inc., Chantilly, VA) at 530 nm. Across 6 assays, the average intra-assay CV was 3.8% and the inter-assay CVs for a pooled serum sample containing 4.31 mg/dL of urea nitrogen was 2.6%.

2.7. Estrous synchronization, breeding, and luteal function

At 81 ± 4 DPP, all heifers were synchronized for ovulation with the 5 d CO-Synch + CIDR protocol that consisted of insertion of an intravaginal progesterone insert (CIDR, Pfizer Animal Health, New York, NY) concurrent with administration of 100 μg of GnRH (Fertagyl, Intervet/Schering-Plough Animal Health, Summit, NJ) at protocol initiation. Five days later, the CIDR was removed and 25 mg of $PGF_{2\alpha}$ (Lutalyse, Pfizer Animal Health, New York, NY) was given at CIDR removal and again 8 h later. At CIDR insert removal, all cows received tail paint (Tell Tail; FIL, 132 Mount Maunganui, New Zealand). At TAI, tail paint scores were assessed (1 = tail paint completely removed; 2 = tail paint partially removed, obvious signs of receiving some rubs from mountings; 3 = no signs of having received mounts, tail paint undisturbed) as an indicator of riding incidences associated with estrous expression before TAI. Seventy-two hours after CIDR removal and initial $PGF_{2\alpha}$ injection, all cows were TAI concurrent with GnRH administration (Fertagyl; 100 μg). All heifers were bred to the same bull by the same technician with all semen procured from a single collection. In addition, transrectal

ultrasound was used to measure the largest cross-section of the dominant ovarian follicle immediately preceding TAI.

At 7 and 14 days following TAI, blood samples were collected for progesterone analysis and handled in an identical manner to that previously outlined. Plasma resulting from theses samples was analyzed in order to indirectly measure CL function. Progesterone quantification was assessed by RIA as previously defined.

Because heifers remained on their respective treatments after TAI, visual AM/PM estrous detection was conducted by trained personnel starting 7 d after TAI and continued until commingling at 32 d after TAI, at which point a bull was placed with the group for the remainder of the breeding season. Any heifers detected in estrus were bred by artificial insemination (AI) based on the AM/PM rule. Pregnancy diagnosis was performed 32 d after TAI using transrectal ultrasonography to determine TAI pregnancy rates. A second pregnancy diagnosis was conducted via transrectal ultrasonography 76 d after TAI to determine 2nd service AI conception rates. Final pregnancy diagnosis was conducted via transrectal ultrasound 45 d after the conclusion of the breeding season to determine breeding season pregnancy rates.

2.8. Pre-weaning progeny performance

Calf BW was measured at birth and again at 64 ± 4 d of age in conjunction with the WSW procedure. At 167 ± 4 d of age, calf BW was measured and calves were provided ad libitum access to creep feed devoid of DDGS (18.2% CP and 1.39 Mcal NE_g/kg on DM basis) until weaning at 191 ± 4 d of age, at which point calf BW was again measured. Creep period intakes and feed efficiency were not collected as all cow-calf pairs were previously commingled and managed as a singular group. However, pre-creep ADG, creep-phase ADG, and overall pre-weaning ADG were calculated.

2.9. Statistical analysis

Differences between treatments for binomial data were analyzed using the GLIMMIX procedure of SAS (SAS Inst. Inc., Cary, NC). The remaining parturition parameters, milk production, milk composition, milk fatty acid profile, and progeny ADG were analyzed using the MIXED procedures of SAS. For dependent variables of interest, the model included the fixed main effect of treatment and animal served as the experimental unit. For all milk related variables, day in lactation was used as a covariate. For progeny related variables, the model also included the fixed effect of sex as well as the treatment × sex interaction; however, in all circumstances, the interaction was not significant ($P > 0.10$) and removed from the model. We do acknowledge that use of animal as the experimental unit when housed in only 2 groups does not allow us to discriminate between effect of treatment and effect of group.

The CORR procedure of SAS was used to determine relationships amongst response variables recorded at parturition. In addition the effect of treatment on duration of anestrous period in females that had resumed estrous activity before the breeding season was evaluated by survival analysis using the Log-Rank method of the LIFETEST procedure of SAS.

Anestrous follicular wave dynamics were analyzed using the MIXED procedures of SAS. The initial model included the fixed main effect of treatment and utilized DPP at follicular wave emergence as a covariate. An ancillary analysis of follicular wave data was also conducted using days before resumption of cyclicity at follicular wave emergence as a covariate.

For performance measurements including heifer BW and BCS as well as progeny BW, the MIXED procedure of SAS for REPEATED measures was utilized. For all variables, the covariance structures compound symmetric, heterogeneous compound symmetric, unstructured, autoregressive order one, heterogeneous autoregressive order one, and ante-dependence were compared. The covariance structure that resulted in the smallest Bayesian information criterion was used for the final analysis. The model included the fixed effects of treatment and day, as well as the appropriate treatment × day interaction. For progeny BW, the model also initially included the main effect of sex and its appropriate interaction with treatment, but was subsequently removed due to insignificance ($P > 0.10$). Animal served as the experimental unit. Simple effects within day were generated using the SLICE function of SAS. For all variables analyzed, a P-value ≤ 0.05 was identified as significant, while a P-value > 0.05 and ≤ 0.10 was identified as a tendency approaching significance.

3. Results

3.1. Heifer performance

As designed, heifer BW and BCS did not differ at the time of treatment initiation ($P \geq 0.60$; Figs. 2 and 3, respectively). Heifer BW did not differ due to dietary treatment ($P = 0.51$), but main effect of day was significant as heifers gained wt throughout the study ($P < 0.001$). In addition, there was a treatment × day interaction for BW (Fig. 2), as heifers in the CON treatment were heavier ($P = 0.002$) than those in the HP treatment at 62 ± 4 d prepartum. Heifer BCS (Fig. 3) was

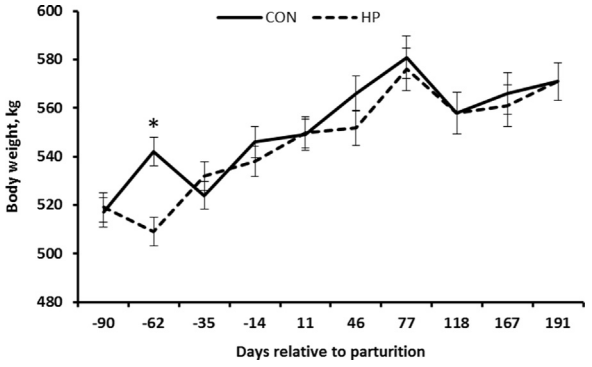

Fig. 2. Effect of treatment (CON = corn silage-based control; HP = dried distiller's grains supplemented as an energy source) on cow BW. A treatment × day interaction ($P < 0.001$) was observed. P-values for treatment and day were 0.51 and < 0.001, respectively. Days on which BW differed between treatments ($P < 0.05$) is indicated with *. Prepartum BW were adjusted for gravid uterine weight (Ferrell et al., 1976).

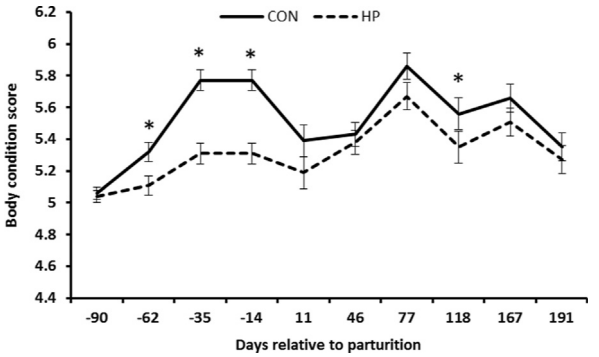

Fig. 3. Effect of treatment (CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source) on cow BCS. Body condition was graded on a scale of 1–9 (1=emaciated, 9=obese; Wagner et al., 1988). A treatment × day interaction ($P<0.001$) was observed. P-values for treatment and day were 0.003 and <0.001, respectively. Days on which BCS differed between treatments ($P<0.05$) is indicated with *.

affected by both treatment ($P=0.003$) and day ($P<0.001$). In addition, a treatment × day interaction was observed for BCS ($P<0.001$; Fig. 3), where CON treatment had greater ($P\leq0.05$) BCS than HP treatment at all prepartum assessments as well as at treatment termination (118 ± 4 DPP).

3.2. Parturition

The proportion of bull calves born did not differ between treatments ($P=0.74$; Table 2). Length of gestation was 2.5 ± 0.7 days longer in heifers on the HP treatment ($P=0.02$), which was accompanied by greater calf birth BW ($P<0.001$), a greater calving difficulty score ($P=0.003$), and an increased rate of dystocia ($P=0.001$) when compared to heifers in the CON treatment (Table 2). In addition, birth BW was positively correlated with gestation length ($P=0.004$; $r=0.34$). Calf vigor shortly after birth and postpartum heifer udder score did not differ due to dietary treatment ($P\geq0.35$). It should also be noted that calf birth BW ($P<0.001$), calving difficulty ($P=0.003$), and rate of dystocia ($P=0.004$) were greater in females that gave birth to bull calves than heifer calves (data not shown). Nonetheless, bull calves tended ($P=0.06$) to have increased vigor scores shortly after birth compared to heifer calves (data not shown).

Increases in birth BW in the current study were positively correlated across treatment to both calving difficulty ($P<0.001$, $r=0.62$) and dystocia ($P<0.001$, $r=0.65$). Despite differences in calving ease and incidence of dystocia between treatments, calf vigor was not correlated with dystocia ($P=0.40$) or calving difficulty ($P=0.87$). Birth BW in the present study, however, tended to be negatively correlated with calf vigor ($P=0.06$; $r=-0.22$).

3.3. Milk

Total milk production measured at 64 ± 4 DPP via WSW did not differ due to dietary treatment ($P=0.86$; Table 3). Likewise, milk protein and milk lactose as a percent of the raw milk sample did not differ due to dietary treatment ($P\geq0.16$; Table 3). In contrast, proportion of milk fat ($P<0.001$) and total milk solids ($P<0.001$) were greater in CON than HP treated heifers (Table 3). Furthermore, when milk composition was included to calculate ECM production, heifers in the CON treatment had greater ECM production per day than heifers in the HP treatment ($P=0.01$). Conversely, HP milk had elevated concentrations of MUN ($P<0.001$) when compared to CON.

Milk fatty acid profiles between treatments are presented in Table 4. Concentrations of all short chain fatty acids (C4:0 to C8:0) and medium chain fatty acids (C10:0 to C15:0) were greater in milk from CON than HP treated heifers ($P<0.001$ for both variables). In contrast, long chain fatty acids ($\geq$C16:0) were greater in the HP than CON treatment ($P<0.001$). Although the concentration of SFA was greater in the CON than HP treatment ($P<0.001$), concentrations of MUFA and PUFA, the ratio of PUFA:SFA, and the concentration of *cis*-9, *trans*-11 CLA were greater in milk from HP than CON treated heifers ($P<0.001$). Furthermore, milk from HP treated heifers had a lower *n*–3:*n*–6 ratio ($P<0.001$) when compared with CON heifers.

Table 2
Effect of yearling beef heifer diet during the third trimester of gestation on calving parameters.

Item	Treatment[a]		SEM[b]	*P*-value[c]
	CON	HP		
Gestation (d)	275.2	277.7	0.73	0.02
Sex of calves (% bulls)	55.3	51.4		0.74
Birth BW (kg)	32.3	36.7	0.64	<0.001
Calving difficulty[d]	1.30	1.81	0.12	0.003
Dystocia (%)[e]	26.3	59.5		0.003
Vigor score[f]	1.27	1.14	0.10	0.35
Udder score[g]	1.14	1.23	0.15	0.55

Parameters.
[a] CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source.
[b] Greater SEM presented ($n=38$ for CON and $n=37$ for HP for gestation, sex, birth BW, calving ease and dystocia; $n=36$ for CON and $n=36$ for HP for vigor and udder scores).
[c] P-values ≤0.05 were considered significant.
[d] Scale of 1–5: 1=no assistance; 2=easy pull; 3=mechanically assisted pull; 4=abnormal presentation; and 5=caesarian-section.
[e] Percentage of females which recorded a calving ease score of 2 or greater.
[f] Scale of 1–4: 1=nursed on own immediately; 2=nursed on own, but slow to start; 3=required assistance to nurse; 4=died shortly after birth.
[g] Scale of 1–4: 1=ideal; 2=not ideal, but calf nursed on own; 3=may require intervention, cull cow with no daughters retained; 4=worst case; cull cow with no daughters retained.

3.4. Ovarian follicular wave characterization

When DPP at wave initiation was included in the statistical model as a covariate, heifers in the HP treatment tended ($P=0.08$) to have longer follicular waves, and maximum diameter attained by the dominant follicle ($P=0.05$) and secondary follicle ($P=0.02$) was greater compared to the CON treatment (Table 5). However, follicle diameter at attainment of dominance, duration of

Table 3
Effect of yearling beef heifer diet fed from 192 days in gestation through 118 ± 4 days in lactation on milk production and composition[a,b].

Item	Treatment[c]		SEM[d]	*P*-value[e]
	CON	HP		
Milk production (kg/d)[f]	8.77	8.86	0.4	0.86
Energy corrected milk production (kg/d)[g]	7.81	6.53	0.35	0.01
Milk protein (%)	3.10	3.19	0.05	0.16
Milk fat (%)	2.36	1.37	0.15	< 0.001
Milk lactose (%)	5.13	5.18	0.03	0.22
Milk total solids (%)	11.5	10.8	0.14	< 0.001
Milk urea N (mg/dL)	7.08	16.35	0.56	< 0.001

[a] Samples collected 64 ± 4 d in milk.
[b] Days in lactation used a covariate in the statistical model.
[c] CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source.
[d] Greater SEM presented (n=33 for CON, n=36 for HP for milk production parameters; n=26 for CON, n=31 for HP for milk composition parameters).
[e] *P*-values ≤0.05 were considered significant.
[f] Measured via 24 h weigh-suckle-weigh procedure that was subdivided into 4, 6-h sampling periods.
[g] Energy corrected milk=(0.3246 × kg milk/d)+(12.86 × kg milk fat/d)+(7.04 × kg milk protein/d).

Table 4
Effect of yearling beef heifer diet fed from 192 days in gestation through 118 ± 4 days in lactation on milk fatty acid composition.

Item	Treatment[a]		SEM[b]	*P*-value[c]
	CON	HP		
(g/100 g of fatty acids)				
C10:0	2.13	1.09	0.09	< 0.001
C12:0	2.83	1.46	0.10	< 0.001
C14:0	10.10	6.02	0.24	< 0.001
C15:0	1.18	0.81	0.02	< 0.001
C16:0	32.30	22.08	0.61	< 0.001
C16:1	2.11	1.41	0.06	< 0.001
C18:0	11.11	13.06	0.49	0.004
C18:1 *cis*-9	26.96	35.03	0.84	< 0.001
C18:1 *trans*-11	1.84	7.59	0.33	< 0.001
C18:2*n*–6	1.58	3.68	0.19	< 0.001
CLA *cis*-9, *trans*-11	0.93	3.37	0.12	< 0.001
Other	6.93	4.46	0.25	< 0.001
Total SFA	64.29	47.60	1.10	< 0.001
Total MUFA	32.09	44.63	0.98	< 0.001
Total PUFA	3.63	7.77	0.25	< 0.001
PUFA:SFA	0.06	0.17	0.007	< 0.001
SCFA[d]	3.74	2.47	0.20	< 0.001
MCFA[e]	17.33	9.97	0.42	< 0.001
LCFA[f]	79.08	87.73	0.53	< 0.001
n–3 fatty acids	0.89	0.37	0.08	< 0.001
n–6 fatty acids	1.80	4.03	0.21	< 0.001
n–3:*n*–6	0.54	0.11	0.03	< 0.001

[a] CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source.
[b] Greater SEM presented (n=26 for CON; n=31 for HP).
[c] *P*-values ≤0.05 were considered significant.
[d] Short chain fatty acids (C4:0 to C8:0; measured but not reported individually).
[e] Medium chain fatty acids (C10:0 to C15:0).
[f] Long chain fatty acids (C16:0 and above).

dominance, and number of antral follicles present on the ovary did not differ ($P \geq 0.11$) between treatments (Table 5).

In an ancillary analysis of follicular wave parameters, days before cyclicity at emergence of the wave was used as a covariate (date not shown). Within this analysis,

Table 5
Effect of yearling beef heifer diet fed during the last trimester of pregnancy through 118 ± 4 days in lactation on follicular wave dynamics when days postpartum at wave emergence was utilized as a covariate in the statistical model.

Item	Treatment[a]		SEM[b]	*P*-value[c]
	CON	HP		
Wavelength (d)	7.45	8.89	0.57	0.08
Dominant follicle diameter (mm)	13.00	15.01	0.72	0.05
Secondary follicle diameter (mm)	9.05	10.67	0.46	0.02
Size at dominance (mm)	9.70	9.14	0.49	0.42
Duration of dominance (d)	4.36	5.78	0.64	0.11
Total follicles				
Day 1 of wave	24.9	24.0	1.82	0.72
Day 2 of wave	21.6	22.8	1.71	0.62
Day 3 of wave	20.9	21.0	1.67	0.97
Average of entire wave	22.5	23.2	1.61	0.75

[a] CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source.
[b] Greater SEM presented (n=10 for CON; n=9 for HP).
[c] *P*-values ≤0.05 were considered significant with $P > 0.05$ and ≤0.10 considered a trend approaching significance.

maximum diameter attained by the dominant follicle (P=0.03) and secondary follicle (P=0.01) in the HP treatment was greater than the CON treatment. However, wavelength, diameter of the dominant follicle at attainment of dominance, duration of dominance, and antral follicle counts did not differ among treatments ($P \geq 0.13$).

3.5. Resumption of cyclicity

The proportion of heifers that resumed estrous cyclicity before estrous synchronization did not differ (P=0.54; Table 6) between treatments. However, the anestrous period for heifers that did resume estrous cycles before estrous synchronization tended to be shorter in HP than CON (P=0.10; Table 6) treatment. Furthermore, the Log-Rank method of survival analysis (Fig. 4) also indicated that HP heifers tended (P=0.10) to resume estrous cycles

Table 6
Effect of yearling beef heifer diet fed during the last trimester of pregnancy through 118 ± 4 days in lactation on resumption of estrous cyclicity and fertility.

Item	Treatment[a]		SEM[b]	*P*-value[c]
	CON	HP		
Anestrous period (d)	63.0	58.4	2.1	0.10
Cyclic at estrous synchronization (%)	90.9 (30/33)	86.1 (31/36)		0.54
Response to GnRH-1[d]	72.7 (24/33)	66.6 (24/36)		0.59
Tail paint score at TAI[e]	1.61	1.53	0.11	0.62
Blood urea N at TAI (mg/dL)	6.63	17.12	0.45	< 0.001
Dominant follicle diameter at TAI (mm)	15.1	15.4	0.29	0.45
Progesterone 7 d after TAI (ng/mL)	3.61	3.73	0.19	0.64
Progesterone 14 d after TAI (ng/mL)	6.99	7.45	0.32	0.30
Pregnancy rates (%)				
TAI	66.7 (22/33)	77.8 (28/36)		0.31
TAI+25[f]	93.9 (31/33)	83.8 (30/36)		0.19
Breeding season	100 (33/33)	97.2 (35/36)		0.98

[a] CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source.
[b] Greater SEM presented (n=33 for CON; n=36 for HP).
[c] P-values ≤0.05 were considered significant.
[d] Proportion of cows that ovulated to the first GnRH administration at CIDR insertion in the 5 day CO-Synch+CIDR protocol.
[e] Tail paint scores were recorded on heifers at timed-artificial insemination, 72 h post-$PGF_{2\alpha}$. 1=no tail paint remaining; 2=some tail paint remaining; 3=tail pain undisturbed.
[f] Proportion of cows diagnosed pregnant to either timed- or 2nd service-artificial insemination.

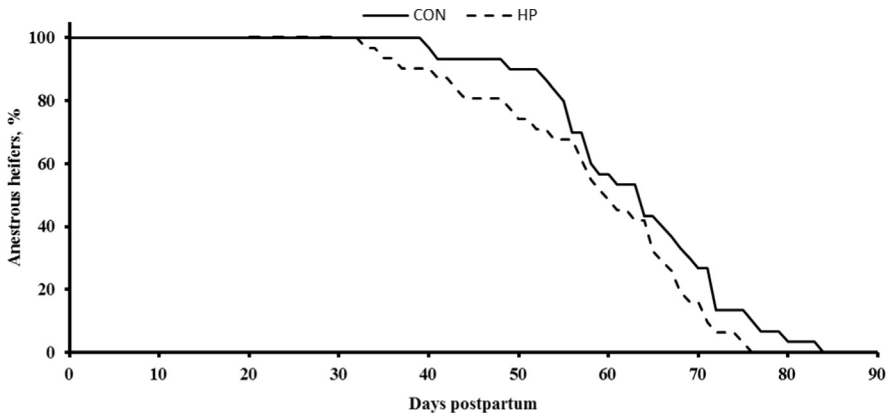

Fig. 4. Effect of treatment (CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source) on length of the anestrous period in heifers that resumed estrous cycles before estrous synchronization. Heifers in the HP treatment tended to resume estrous cycles (P=0.10) earlier during the postpartum period than CON treated heifers.

earlier in the postpartum period than heifers in the CON treatment.

3.6. Blood urea nitrogen

There was a significant ($P < 0.001$) treatment, day, and treatment × day interaction detected for BUN concentrations (Fig. 5). At all three sampling times, BUN concentrations were higher in HP compared to CON, but the magnitude of difference during the postpartum sampling times were lesser than those observed during the prepartum period.

3.7. Estrous synchronization, breeding, and luteal function

Dietary treatment did not affect tail paint scores (P=0.62) or diameter of the dominant follicle at TAI (P=0.45). Likewise, proportion of heifers pregnant to TAI

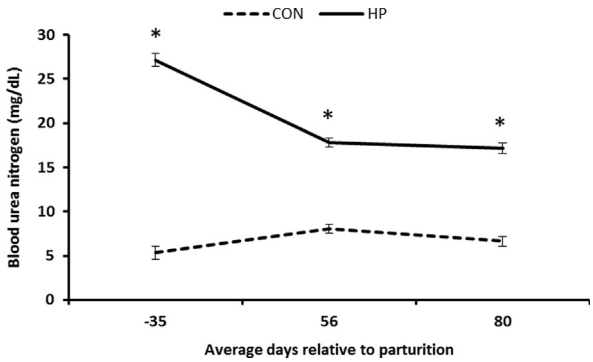

Fig. 5. Effect of treatment (CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source) on circulating blood urea nitrogen (BUN) concentrations. There was a significant effect of treatment ($P < 0.001$) and day ($P < 0.001$) on BUN, and a treatment × day interaction ($P < 0.001$) was also observed. Days on which BUN differed between treatments ($P < 0.05$) is indicated with *.

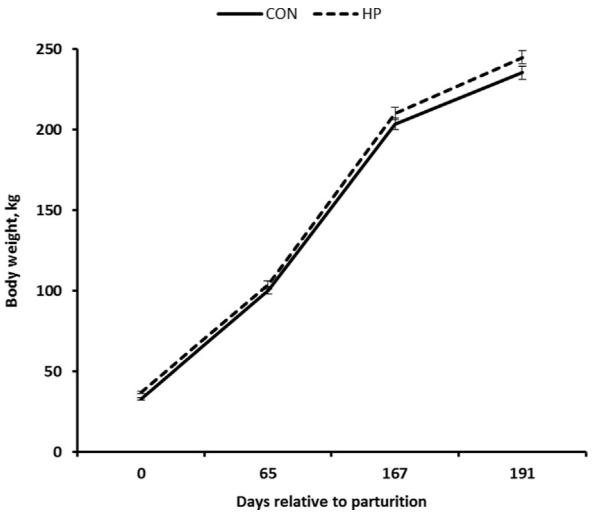

Fig. 6. Effect of treatment (CON=corn silage-based control; HP=dried distiller's grains supplemented as an energy source) on pre-weaning progeny growth. *P*-values for treatment and day were both <0.001, respectively. Steer progeny were heavier than heifer progeny throughout the pre-weaning period ($P<0.001$). There were no significant treatment × day or treatment × sex interactions ($P>0.10$).

($P=0.31$), concentrations of progesterone at 7 or 14 d after TAI ($P\geq 0.30$), 1st and 2nd service AI pregnancy rates, nor overall breeding season pregnancy rates ($P\geq 0.98$) were affected by dietary treatment (Table 6).

3.8. Pre-weaning progeny performance

Body weight of progeny born to heifers in the HP treatment was greater throughout the pre-weaning period when compared to progeny of CON heifers ($P<0.001$; Fig. 6). In addition, bull calves were heavier than heifer calves ($P<0.001$; data not shown) across treatments and day. Progeny ADG during the pre-creep period did not differ due to dietary treatment ($P=0.20$). In contrast, during the 24-d creep period (167 to 191 ± 4 d of age) immediately prior to weaning, HP progeny had a greater ADG ($P=0.02$; 1.34 vs. 1.21 ± 0.04 for HP and CON, respectively), which translated to a tendency for increased ADG over the duration of the pre-weaning phase ($P=0.06$; 1.09 vs. 1.06 ± 0.02 for HP and CON, respectively) when compared to CON progeny.

4. Discussion

The objective of this study was to assess differences in various performance parameters of late gestation and early lactation heifers and their progeny when late gestation and lactation diets significantly exceeded CP requirements. This was accomplished by feeding DDGS, a commonly available corn by-product, as a primary dietary energy source. It must be noted that corresponding with this greater CP content in females fed DDGS, the chemical composition of DDGS also resulted in increased dietary fat content. However, in order to eliminate this factor and the potential difference in animal BW gains as a confounding factor, initial diets were formulated, based on chemical composition of individual ingredients, to provide similar daily megacalories of NE_g and maintain similar BW between treatments throughout the study. It should be noted that cows in both treatments obtained the projected ADG during both the pre- and postpartum periods. With the exception of -62 ± 4 d relative to parturition, BW remained similar between treatments throughout the treatment period. The difference in BW on -62 ± 4 d is most likely attributed to differences in gut fill, as heifers were not allowed a dietary adjustment period to normalize rumen fill prior to trial initiation.

Although BW was similar between treatments throughout the majority of the study, prepartum BCS was greater in CON than HP treated heifers during the prepartum period and at treatment termination. Even though there may be several contributing factors to these differences in BCS between treatments, we hypothesize that the most likely reason is associated with dietary fat source and subsequent site of fat deposition. Feeding DDGS has been reported to shift site of fat deposition from subcutaneous to visceral (Depenbusch et al., 2009), potentially through protection of unsaturated fatty acids from ruminal biohydrogenation in DDGS-based diets (Vander Pol et al., 2009). Nonetheless, BCS is a subjective measurement of subcutaneous fat depots, thus a difference in lipid metabolism and potential changes in location of accretion in the current study could account for differences in BCS noted between treatments.

The increase in birth BW of calves born to HP treated heifers concurs with recent studies that reported increased birth BW (Radunz et al. 2010, 2012; Wilson et al., 2012) in calves born to cows fed DDGS during the last trimester of gestation. In the current study, birth BW of calves was positively correlated with gestation length. Such a correlation could suggest that the increased gestation length in HP treated heifers is responsible for the greater birth BW. However, the 2.1 d difference in gestation length between treatments in the current study only contributed approximately 1 kg of the 4.4 kg difference in birth BW observed between treatments in the current study based on fetal growth estimates calculated from Ferrell et al. (1976) equations. The mechanism(s) by which increased CP and/or fat content of the diet increases birth BW have not yet been elucidated.

We acknowledge that the rate of dystocia in the current study is relatively high; however, results are comparable to primiparous heifers studied by Notter et al. (1978). Although it is conceivable that increased dystocia in the HP treated heifers could be a result of the proposed increase in visceral fat deposition previously discussed, internal fat measurements were not conducted in the current study, and thus future studies may be warranted to validate such a claim. Although birth BW, calving ease, and dystocia were greater in progeny born to HP treated heifers, calf vigor was not affected, nor was calf vigor correlated with dystocia or calving difficulty. This is in contrast to Colburn et al. (1997) who reported poorer calf vigor scores with concurrent increases in calving difficulty.

Given that the energy provided in the current study met the requirements for first-parity heifers and that

the genetic background of the heifers was similar, the observation of similar total milk production between treatments was expected. Limited research has been conducted investigating the consequences of feeding DDGS on milk production in beef cows. Radunz et al. (2010) reported that feeding DDGS at 66.5% of DMI did not affect milk production when compared to hay alone or a diet of 60% corn and 27.5% hay. However, Shike et al. (2009) reported suppression in milk production in cows fed a diet of 55% DDGS when compared with cows fed a diet containing 57% corn gluten feed. Although milk production did not differ between treatments, milk composition varied. Differences in milk composition data in the current study are likely a function of associative effects between concentrate and roughage within individual diets. We acknowledge that milk fat values are considerably less than expected, which could be due to incomplete emptying of the quarter at sampling. Milk fat concentrations in the current study, however, were similar to those reported by Winterholler et al. (2012) in which beef cows were supplemented with up to 4 kg/d of DDGS. It is likely that added dietary fat found in the HP treatment may have had a negative associative effect on fiber digestion, leading to decreased acetate production and suppressed milk fat (reviewed by Coppock and Wilks, 1991). The decrease in milk fat coupled with a lack of differences in total milk production led to a decrease in ECM production in HP treated heifers when compared to CON. This could be of particular concern as suppression in ECM production has the potential to result in decreased pre-weaning progeny performance.

Due to the prevalence of unsaturated fatty acids in DDGS, it is not surprising that there was a shift in milk fatty acid profile toward greater concentrations of MUFA and PUFA in HP treated heifers when compared to CON. The shift in composition was much more pronounced than that reported by Schingoethe et al. (1999), where wet distillers grains were included at a lower concentration within the diet (31.2% of diet DM). Milk from HP treated cows in the current study had a nearly 3.5-fold increase in *cis*-9, *trans*-11 CLA concentration when compared to CON. Increases in CLA content of the milk associated with feeding DDGS is not unprecedented Leonardi et al. (2005) and Anderson et al. (2006) both reported increases in *cis*-9, *trans*-11 CLA in dairy cows fed DDGS.

In cattle, ovarian follicular development resumes within days after calving (Savio et al., 1990; Murphy et al., 1990; McDougall et al., 1995; Stagg et al., 1995). Each successive follicular wave is longer in duration and the dominant follicle achieves a greater diameter before atresia (Murphy et al., 1990) until ovulation is achieved. In the current study, when DPP was used as a covariate in the statistical model, the HP treatment tended to have a follicular wave of greater duration, and the dominant and secondary follicle reached a greater maximum diameter than the CON treatment. Because there was no difference between treatments in DPP at the beginning of the characterization period, such observations could indicate that feeding DDGS either directly enhances follicular growth patterns or was hastening resumption of estrous cyclicity, thus advancing follicular growth. Hence, it was unclear if these differences were attributed directly to dietary treatment or if these differences were an indirect effect of the number of days before estrous cyclicity at wave initiation. Therefore, an ancillary analysis was conducted using days before resumption of cyclicity as a covariate. When the secondary analysis was conducted, the tendency in wavelength difference between treatments was nullified, indicating that treatment was likely regulating duration of anestrous and not directly regulating follicle wavelength. Nonetheless, in the ancillary analysis both maximum diameter of the dominant and secondary follicle remained greater in the HP than CON treatment. These data indicate that differences in follicle diameter were a direct effect of dietary treatment and not merely a function of time relative to resumption of cyclicity.

It has been demonstrated that dietary energy concentration can impact dominant follicle growth in primiparous (Ciccioli et al., 2003) and mature (Grimard et al., 1995) beef cows. While it should not be ignored that reduced ECM in the HP treatment may have resulted in more energy available to stimulate follicular growth; we speculate that the increased follicular growth in HP fed heifers was the result of either increased dietary fat, excessive protein, or a synergistic effect of both of these components. Bilby et al. (2006) reported that increasing PUFA concentrations in the diet increases the size of the ovulatory dominant follicle in lactating dairy cows, which may be a result of increased plasma IGF-1 and cholesterol due to dietary linoleic acid (Robinson et al., 2002). Alternatively, Lents et al. (2008) reported that postpartum beef cows grazing dormant forage tended to have larger ovulatory follicles when supplemented with CP, which is supported by recent data collected in our laboratory (Gunn et al., 2013) where excess dietary protein rich in rumen undegradable protein fed at 150% of NRC requirements increased ovulatory ovarian follicle growth in nonpregnant, nonlactating beef cows.

Given that all nutrient requirements were met, energy provided was similar between treatments, and DPP was extended prior to estrous synchronization, it was not surprising that such a large proportion of heifers were determined to have resumed estrous cyclicity at the initiation of estrous synchronization. Bolze et al. (1985) reported a shorter postpartum interval for beef cows fed 150% of NRC CP requirements when compared to cows fed 100% of NRC CP requirements during the last 112 d prior to calving. Moreover, the tendency in decreased anestrous period in the HP treatments is supported by alterations in follicular patterns as previously discussed and by Jones et al. (2008) who reported an earlier resumption of cyclicity in dairy cows supplemented with greater concentrations of rumen protected linoleic acid. In addition to PUFA effects on the postpartum interval, a decreased anestrous period associated with overfeeding protein during late gestation is not unprecedented. As previously discussed, excess CP intake, or the increase in ruminally protected unsaturated fatty acid intake provided by the HP treatment, or both, may have the potential to increase IGF-1, which has previously been shown to be positively correlated to a decreased postpartum interval (Patton et al., 2007).

Increased BUN concentrations in the HP treatment when compared to CON were expected, and these differences are consistent with those of Radunz et al. (2010). As protein intake increased in the postpartum diets of the CON treatment, it was not surprising that CON BUN concentrations increased. However, increased demands of CP for continued growth of the dam and milk production, coupled with urea excretion in the milk, likely explains the suppression in postpartum BUN within the HP treatment and therefore the reduction in BUN from 56 to 80 DPP within the CON treatment. More importantly, though, is the role that increased BUN concentrations may play in the ability for a cow to conceive and/or maintain a pregnancy. In a review of dietary protein in dairy cattle, Butler (1998) concluded that BUN concentrations exceeding 19 to 20 mg/dL are associated with decreased fertility. Although BUN concentrations at TAI for HP (17.12 ± 0.56 mg/dL) were approximately 2.5 times greater than that of CON (6.63 ± 0.25 mg/dL), they were not greater than the aforementioned concentration associated with suppressed fertility in dairy cows.

Although not statistically significant, the 16.6% increase in TAI pregnancy rate of the HP treatment compared to CON was surprising. It is recognized that statistical power was limited within this study for assessment of binary variables; however, these data are complementary to other studies within our laboratory that reported a numerical increase in TAI pregnancy rate in cows fed DDGS. Gunn et al. (2014) reported an 18% increase (50.5 vs. 42.2%) and Shee et al. (2012) reported a 69% increase (81.5 vs. 48.1%) in TAI pregnancy rates for DDGS fed cows compared with control cows fed a similar amount of energy on a daily basis. Despite elevated dietary protein concentrations typically noted with DDGS-based diets, to our knowledge, suppression of pregnancy rates in beef females fed DDGS at low to moderate levels within the diet has not been reported. One possible explanation for this is that increased PUFAs in the diet may improve early embryo quality (Thangavelu et al., 2007; Cerri et al., 2009). Alternatively, Berardinelli et al. (2001) reported that day 5 embryos recovered from ewes fed a diet containing 200% of NRC requirements for protein had a greater number of cells per embryo when compared with ewes fed 100% of NRC CP requirements, indicating improved embryonic development. One other possible explanation is that *n*–3 fatty acids derived from a diet rich in DDGS may reduce prostaglandin production and secretion (Mattos et al., 2002, 2003, 2004), allowing for a greater rate of embryo survival. Nonetheless, given the limited number of observations in this and other studies within our laboratory, further studies are warranted to fully elucidate the effects of CP and DDGS on fertility and the mechanisms by which potential differences are mediated.

Increased BW at birth for progeny of HP treated heifers when compared with CON treated offspring was maintained through 167 ± 4 d of age. This was somewhat unexpected because energy corrected milk production was lower in HP treated heifers, suggesting that CON progeny should have narrowed the BW margin during this time period. These data, coupled with the increased ADG of HP progeny during the 24-d creep period immediately before weaning, indicate that HP progeny may have been more efficient during the pre-weaning period. It should be noted, however, that intake of maternal diets by calves during the neonatal treatment period in the current study was possible, and thus could have contributed to differences in BW. Data of Larson et al. (2009), however, support the theory of differences in performance, as offspring born to dams supplemented protein during the last trimester of gestation (although not different in BW at birth) had a tendency for increased adjusted 205-d weaning wt when compared to the progeny of non-supplemented dams. Alternatively, it is conceivable that differences in maternal milk composition altered calf growth through lactational programming (Soberon and Van Amburgh, 2012). Because PUFA concentrations were greater in the milk of HP than CON cows in the current study, it is possible that differences in pre-weaning progeny growth were mediated through altered IGF-1 concentrations (Robinson et al., 2002).

In summary, feeding excess CP and dietary fat in the form of DDGS as an energy source during late gestation and early lactation to first-parity heifers resulted in increased birth BW and rate of dystocia. Although total milk production was not altered, ECM production was less in HP females. In addition, HP females had greater dominant follicle growth during the postpartum anestrous period and marginally hastened resumption of estrous cycles. Furthermore, heifers fed DDGS had altered BUN concentrations throughout the treatment period. Nonetheless, feeding DDGS as a primary energy source did not detrimentally impact AI pregnancy rates as hypothesized. However, calves from HP treated dams were heavier at weaning when compared to CON progeny. Although HP treated dams had an increased incidence of dystocia, feeding excess dietary CP and increasing dietary fat via DDGS did not negatively impact cow growth and reproductive performance or progeny performance. However, further research should be directed at what chemical components of DDGS are the causes of differences in reproductive function and progeny growth outlined in this study.

Conflicts of Interest

We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome.

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