

Effects of reduced *in utero* and post-weaning nutrition on milk yield and composition in primiparous beef cows

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(Received 24 November 2015; Accepted 26 May 2016; First published online 30 June 2016)

Development and long-term retention of replacement beef females in a semi-arid environment are of a major concern for extensive livestock producers. Furthermore, the demand of not only producing a thriving, healthy calf, but having sufficient milk to support that first calf is essential. To address this issue, we conducted a 3-year study measuring milk production and milk constituent yields in primiparous beef heifers (n = 48; 16/year reared under two different feeding regimens) raising steer calves. Cows received 1.8 or 1.2 kg/day winter supplementation for ~80 day before parturition and their heifer calves were then randomly assigned to heifer development treatments that provided ad libitum (AL) or 80% (less than ad libitum (LAL)) of ad libitum feed post weaning. Heifers developed on the AL treatment also received 1.8 kg/day winter supplementation for life, whereas heifers developed on the LAL treatment received 1.2 kg/day winter supplementation for life. Milk production of primiparous cows was measured with a portable milking machine every other week from days 27 to 125 postpartum. Milk yield for the 125-day lactation period was calculated from area under the lactation curve approximated by trapezoidal summation. The ANOVA model included in utero winter nutrition, post-weaning heifer development treatment, year and their interaction. Heifers subjected to the AL treatment reached peak milk yield ~12.3 day later (P = 0.02) than heifers receiving LAL treatment. In addition, an in utero nutrition \times post-weaning heifer treatment \times year interaction existed (P \leq 0.04) for milk peak yield, average daily milk yield (kg/day) and nutrient composition (protein, lactose, fat, solids non-fat, g/day). These interactions manifest as changes in magnitude and rank across the 3 years of the study. Livestock production in extensive environments is subject to variations in seasonal precipitation patterns and quality and quantity of grazeable forage and these fluctuations have a large impact on milk yield. In summary, the gestational nutritional environment of a heifer's mother may interact with the heifer's nutrient consumption during post-weaning growth and the current year to trigger variation in year-to-year milk production.

Keywords: heifer development, milk and constituent yield, primiparous beef cow

Implications

Raising livestock in an extensive rangeland environment relies heavily on the nutritive quality of grazeable forage. However, livestock producers in arid and semi-arid environments often encounter times of the year when grazeable forage quality is inadequate to meet animal requirements. Therefore, producers often resort to supplementing livestock with harvested feeds, which ultimately increases cost of production. A challenge for researchers is to provide producers with new and acceptable approaches to sustain optimal levels of production while ensuring economic feasibility. Present research suggests that obtaining an optimal level of additional feed inputs rather than maximizing intake has a similar impact on primiparous milk production. Therefore, by

not maximizing but reducing the amount of supplementary harvested feeds during times of diminished forage quality lowers cost of production and maintains production goals.

In Northern Great Plains, USA, a common practice is to supply harvested feedstuffs during the late gestation period to cows grazing senesced rangelands due to the low quality of forage available. The question then arises as to what extent this feeding paradigm has on subsequent growth and development of offspring. Any time harvested feeds are used to augment the nutritional environment in which grazing livestock are being raised, there is a associated, direct cost to production. This associated cost of providing harvested feedstuffs can have detrimental impacts on extensive livestock operations if less than desirable production goals are not achieved. This includes calving ease, calf vigor,

Introduction

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milking ability of the dam, post-calving growth rate of calf, timely resumption of estrus for the dam and how post-weaning treatment of replacement females' impact sustainability of extensive livestock operations. Pre-pubertal growth rate of heifers has been found to be positively related to their subsequent milk production (Buskirk et al., 1996; Sejrsen et al., 2000). Further, cows with similar genetic potential for mature weight may differ in production efficiency depending on their milk yield (Montano-Bermudez and Nielsen, 1990). Calves born to dams developed on 80% ad libitum feed after weaning and provided a reduced level of supplemental feed during winter before calving weighed less at birth and weaning than contemporary calves from dams that were provided with ad libitum feed during development and(or) before calving (Roberts et al., 2009b). One plausible explanation for this result is that milk yield or composition was affected by the imposed nutritional regimens during post weaning or in utero development. Thus our objective was to evaluate effects of the previously established nutritional paradigms (post-weaning and in utero nutrition) on yield and composition of milk.

Material and methods

Study location and environment

This study was conducted at the USDA-ARS Fort Keogh Livestock and Range Research Laboratory (LARRL) located ~1.6 km west of Miles City, MT (46°22'N 105°5'W) from April 2009 through August 2011. LARRL Institutional Animal Care and Use Committee approved all animal handling and experimental procedures used in this current study.

The LARRL encompasses 22 500 ha and has an average elevation of 730 m. Average daily temperatures range from

-12°C in January to 24°C in July with daily maximum temperatures occasionally >37°C during summer and daily minimum temperature occasionally dropping < - 40°C during winter. Winter mean temperatures during the study were similar to the long-term average (Figure 1). Average annual precipitation is ~340 mm and occurs primarily between April and September (Figure 1). Precipitation was 288 mm in 2008, 260 mm in 2009, 452 mm in 2010 and 481 mm in 2011. Thus, the year before initiation of the study and the 1st year of the study had less than average precipitation, whereas precipitation during the last 2 years of the study was greater than the long-term average. May precipitation was greater than the long-term average in 2010 and 2011 by 74 and 179 mm, respectively. Predominant grasses at this site include grama (Bouteloua), needlegrass (Hesperostipa) and wheatgrass (Pascopyrum; Küchler, 1964). Average standing crop of forage produced annually at the study site is $870 \pm 14 \,\mathrm{kg/ha}$ (Grings et al., 2005). Stocking rate was 1.27 ha/animal unit month and the quantity of forage available in the 53.8 ha pasture during 128 days that primiparous cows were on trial was in excess of cattle needs. Forage characteristics are described in Table 1 and indicate that CP concentrations typically are elevated in early to mid-spring and taper off as summer advances and plants senesce. However, CP concentrations during the period when primiparous heifers were nursing met or exceeded requirements (NRC, 2000) throughout the 128 day study when milk production was measured.

Two ruminally cannulated cows grazed with experimental suckled primiparous cows throughout the study. Samples of grazed forage extrusa were collected and analyzed to estimate diet quality and describe nutrient composition of forages grazed. Diet extrusa samples were collected in April, May and June of 2009 and 2010. Diet extrusa was not

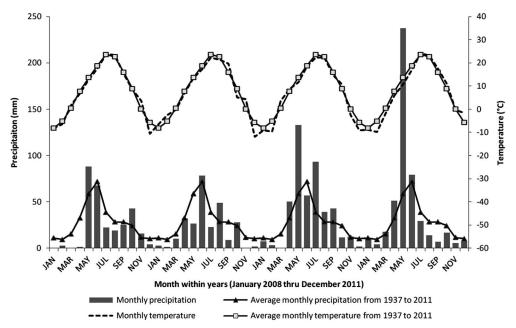


Figure 1 Precipitation and temperature measures during 2008 and 2011 along with 74-year averages at Miles City, MT. (National Oceanic and Atmospheric Administration, 2008–11).

Table 1 Forage characteristics of rumen extrusa samples collected in April, May and June during the time when first parity heifers were grazing experimental pasture

		2009			2010	
Items	April	May	June	April	May	June
% DM						
DM	88.63	89.99	90.74	89.71	89.52	91.18
CP	15.70	13.70	8.53	13.00	19.80	9.70
ADF	34.80	36.90	40.00	42.70	33.80	39.00
TDN	62.90	60.50	56.90	53.90	64.00	58.10
S	0.19	0.19	0.14	0.19	0.25	0.14
Р	0.71	0.52	0.29	0.41	0.57	0.42
K	2.21	1.50	1.34	0.88	2.08	1.15
Mg	0.20	0.17	0.15	0.11	0.21	0.13
Ca	0.73	0.50	0.44	0.38	0.54	0.48
Na	2.57	2.12	1.50	2.41	1.98	1.47
ppm						
Fe	389.00	991.00	274.00	902.00	594.00	205.00
Mn	111.00	106.00	72.00	103.00	110.00	78.30
Cu	10.30	6.00	4.00	4.50	8.00	4.60
Zn	31.60	30.70	23.10	29.90	34.50	25.20

DM = dry matter; TDN = total digestible nutrients.

collected in 2011 due to persistence of precipitation and inability to keep cannulated animals from consuming standing water that might bias nutrient analysis of the ruminally recovered samples. Diet samples were obtained following complete removal of rumen contents and immediately sponge-dried to remove any residual moisture as described by Lesperance *et al.* (1960). Briefly, rumen evacuated cows were released into experimental pastures and allowed to graze for 45 to 60 min. After the grazing period, grazed forage extrusa was removed from the rumen and thoroughly mixed and individually frozen at -20° C, lyophilized, ground to pass a 2 mm screen and stored at -20° C until analysis by a commercial laboratory (Midwest Laboratories Inc. Omaha, NE, USA).

Herd management

Primiparous cows (*n* = 48) from the stable composite gene population (1/2 Red Angus, 1/4 Charolais, 1/4 Tarentaise) described by Newman *et al.* (1993a and 1993b) nursing steer calves were used in this study. Management of this population is such that dams receive a lifetime treatment of either adequate (ADEQ) or marginal (MARG) winter supplementation consisting of alfalfa hay (1.8 or 1.2 kg/day) for 80 day before parturition (Roberts *et al.*, 2010). In Brief, at weaning heifer calves are removed from their dams and relocated to dry lots and developed in pens containing Calan gates (American Calan, Northwood, NH, USA) to facilitate individual feeding. Post-weaning heifer developmental diets consisted of either *ad libitum* (AL) or 80% of *ad libitum* (less than *ad libitum* (LAL)) feed availability throughout a 140-day development period. Subsequent adjustments in the amount

to feed offered to heifers on the LAL treatment were calculated every 28-day using the following formula:

 $LAL = [0.80 \times (mean \, BW \, of \, restricted/mean \, BW \, control) \\ \times mean \, daily \, feed \, intake \, (as-fed \, basis) \, of \, controls \, over \\ the \, 28-day \, period].$

Following this 140-day developmental treatment period (just before breeding), developed heifers were commingled and managed similarly through their first breeding season. Upon pregnancy confirmation, the now primiparous cows were assigned to lifetime fall/winter treatments. The now primiparous cows, which had received the AL post-weaning development treatment, were supplemented at the ADEQ level (1.8 kg/day) and the primiparous cows that received the LAL post-weaning development treatment were supplemented at the MARG (1.2 kg/day) level of fall/winter supplementation. Just before calving, primiparous cows were again commingled and managed similarly throughout the year until the following year when they were once again separated into their respective fall/winter treatments. Description of experimental design and herd management has previously been reported (Roberts et al., 2007 and 2009a; Waterman et al., 2011).

Experimental animals

In total, 16 2-year old, primiparous cows nursing steer calves were selected from the aforementioned paradigm in each of the 3 years studied. Primiparous cows were chosen to provide equal subclass numbers for *in utero* nutrition for 80 days before parturition (ADEQ and MARG, n=24 respectively) and post-weaning heifer development treatment (AL and LAL, n=25 and 23, respectively) and for having steer calves of similar age.

Measurements

Milk collection began ~27 days (earliest date to acquire selection criteria previously mentioned) after parturition (average 16 April each year) and continued at 14-day intervals for 112 days using a technique previously described (Waterman et al., 2006). In brief, on the day of each milking, cows were gathered from pasture, calves were removed and cows were administered an i.m. injection of oxytocin (40 USP; Bayer HealthCare, LLC. Shawnee Mission, KS, USA) 5 min before milking to facilitate milk letdown (Beal et al., 1990). Heifers were milked using a portable milking machine (SuperKart, Coburn Company Inc., Whitewater, WI, USA). Milk collected from the initial milking was discarded. Heifers were kept separate from calves for ~4 h (exact time (hundredths of a second) was recorded and used in calculation), and then milked a second time using the same procedures. Weight of milk from the second milking was recorded, and an aliquot (20 ml) was retained for analysis of milk protein, lactose, fat, solids non-fat and milk urea N by Rocky Mountain DHIA (Logan, UT, USA). Daily milk yield was estimated from the 4h milk production yield (kg/day) and composition (g/day) was calculated by multiplying constituent concentration by daily milk production (Appeddu *et al.*, 1997; Waterman *et al.*, 2006).

Statistical analysis

Milk yield for lactation period, peak milk yield, day to peak milk yield and milk yield from peak to final milk measure were determined using GraphPad Prism software (GraphPad Software Inc., San Diego, CA, USA). Trapezoidal summation was used to determine total lactation period milk yield and milk yield from peak to last milking. Persistency of milk production was calculated from day to peak yield to final milk measure for each cow using the following equation (DHI, 2015). The denominator days between tests had a mean of 78 ± 3.1 day and ranged from 14 to 98 day.

$$1 - \left(\frac{\frac{(\text{Milk yield } (\text{kg}) \text{ at peak} - \text{Milk yield } (\text{kg})}{30 \text{ days}}}{\frac{30 \text{ days}}{\text{Days between tests}}}\right) \times 100$$

Milk yield and constituents (protein, lactose, fat, solids non-fat g/day and milk urea N, mg/100 ml) were analyzed by fitting a repeated measures mixed model using the MIXED procedure of SAS 9.4 (SAS Institute Inc., Cary, NC, USA). The model included fixed effects of in utero nutrition, and post-weaning heifer development treatments, day of milking and all interactions for fixed effects. Animal and year effects were fitted as random effects. The covariance of the repeated measures factor was modeled with the first-order auto regressive covariance structure AR(1). Covariates used in the analysis of milk yield and composition included calf birth weight and days in milk. Statistical significance was set at $P \le 0.05$. The Bonferroni method was used to adjust *P*-values for multiple comparisons. Average milk yields and composition were analyzed using the MIXED procedure of SAS (SAS Institute Inc.). The model included fixed effects of in utero nutrition, and post-weaning heifer development treatments, year and all interactions. Data were fitted to the following statistical model:

$$y_{ijklm} = \mu + A_{i(jkl)} + \gamma_l + \alpha_j + \beta_k + \rho_m + \alpha\beta_{jk} + \alpha\rho_{jm}$$
$$+ \beta\rho_{km} + \alpha\beta\rho_{jkm} + bw_{ijkl} + dim_{ijkl} + e_{ijklm}$$

Where Y is the observation, μ is the overall mean, $A_{i(jkl)}$ the ith animal effect within each treatment and year, γ_1 the lth year effect, α_j the jth in utero treatment, β_k the kth post-weaning treatments, ρ_m the mth day effect (repeated measures factor and is a fixed effect), and ethe associated error. The denominator DF for significant testing were adjusted using the Kenward–Rodger approximation. The REPEATED statement included collection date with the subject cow by (in $utero \times$ heifer development treatment) using an autoregressive covariance structure. Covariates used in the analysis of milk yield and composition included calf birth weight, and days in milk. Statistical significance was set at

 $P \le 0.05$. The Bonferroni method was used to adjust *P*-values for multiple comparisons (SAS Institute, 2010).

Results and discussion

In utero and post-weaning heifer development milk production

Days postpartum to peak milk production was affected (P = 0.02) by post-weaning heifer development treatment (Table 2). Primiparous cows which received the postweaning AL development treatment reached peak milk production ~12.3 days later (7.6 weeks postpartum), whereas heifers receiving the LAL post-weaning development treatment achieved peak lactation at ~5.9 weeks postpartum. Interval to peak milk yield has been shown to range from 3 to 7 weeks postpartum in beef cows depending on ecophysiological environment, season of calving and nutritional environment (Wood, 1972; Totusek et al., 1973; Grings et al., 2008). The 12.3 days less to peak milk production in primiparous cows developed on reduced heifer development diets may partially be explained by reduced BW postpartum in cows developed on the LAL treatment (361 v. 336 ± 5.3 kg for AL and LAL, respectively). In the present study, it is not possible to differentiate whether the decreased BW postpartum was a carry-over effect of LAL during heifer development or the MARG diet that primiparous cows consumed just before their first parturition that resulted in the 12.3 day delay to peak milk yield.

Effects of in utero nutrition and post-weaning heifer development treatment for total milk area under curve (AUC) were not different ($P \ge 0.31$). However, when milk AUC from peak milk production to final milk measure collected in this study, a post-weaning heifer development treatment by year effect (P = 0.05) was observed. Thus, indicating that primiparous cows that received the post-weaning LAL development treatment had greater milk yield in the first 2 years from peak to final milking and lower milk yield the last year compared with AL developed heifers. Looking at precipitation patterns across the years (Figure 1), milk yield from peak to final milking decreased as precipitation increased and in year 3 when precipitation was the greatest during the lactation period heifers developed on LAL produced the least amount of milk. This indicates that primiparous cows developed on LAL may be more susceptible to changes in environmental conditions than their primiparous cohorts developed on AL.

However, no differences (P = 0.79) for milk persistency measures from peak milk to final milking were measured for *in utero* nutrition (82.2 v. 81.0 \pm 2.94% for ADEQ and MARG, respectively) or post-weaning heifer development treatment (P = 0.31; 79.5 v. 83.7 \pm 2.84% for AL and LAL, respectively).

An *in utero* nutrition \times post-weaning heifer development treatment \times year interaction existed for peak milk yield (P=0.01; Figure 2). This interaction resulted from changes in rank across years. Primiparous cows that received ADEQ *in utero* nutrition and developed on AL had the least variability in peak milk yield across the 3 years of the study (5.6 to 6.7 kg/day) whereas ADEQ \times LAL had the lowest peak yield

Least square means for milk yield and constituents on in utero nutrition (cow) and post-weaning heifer development (heifer) over an 18-week period Table 2

	In utero TRT*	2 TRT*	•	Heifer TRT⁺	TRT⁺	'•		Year						Р	P-value		
Items	ADEQ	ADEQ MARG SEM AL LAL SEM 2009 2010 2011	SEM	AL	LAL	SEM	2009	2010	2011	SEM	<i>In utero</i> TRT	Heifer TRT	Year	<i>In utero</i> TRT × Heifer TRT	<i>In utero</i> TRT×year	Heifer TRT × year	<i>In utero</i> TRT×heifer TRT×year
u	24 24	24		25 23	23		16 16		16								
Milk production																	
Days (milk peak)	47.4	47.1	3.72	53.4	41.1	3.58	53.1	8.09	27.8 8.77	8.77	96.0	0.02	<0.01	0.38	0.39	0.16	0.65
Milk AUC (kg \times 98 days)	435.3	435.3 434.8 21.80 450.1 416.9 21.0	21.80	450.1	416.9	21.0	610.3 4	101.8	401.8 293.0 51.46	51.46	0.98	0.31	<0.01	96.0	0.23	08'0	0.12
Milk AUC from peak	316.3	322.9	26.48								0.87	0.18	0.14	0.42	0.21	0.05	0.17
AL						-	350.6 237.2 297.1 62.0	237.2	297.1	52.0				-	-	-	-
LAL							523.0 260.1 249.6 77.77	260.1	249.6 7	77.77						-	-
Milk urea N, (mg/100 ml) 9.6 9.8 0.29 10.0 9.4 0.29	9.6	9.8	0.29	10.0	9.4	0.29	8.7 9.1 11.3 0.36	9.1	11.3	98'0	0.51	0.34	<0.01	0.75	0.85	0.51	0.36
ADEO - Adoutington TDT - transformet MADC - marginal AI - ad libitation 101	AM .taoat.	- T	-leainae	- IV	l lihitum:		loce than	tidil be) I V	7	Chair when soft is it is a with the many						

ADEQ = adequate; TRT = treatment; MARG = marginal; AL = ad libitum; LAL = less than ad libitum; AUC = area under curve. *Cows whom received in utero nutrition of ADEQ (1.8 kg/day) or MARG (1.2 kg/day) levels of winter supplement.

Comparison of heifers fed ad libitum or 80% of ad libitum fed (LAL; adjusted to a common BW) for 140-day post weaning and fed the equivalent of either 1.8 kg/day (AL) or 1.2 kg/day (LAL) supplement during the winter receding calving. Milk AUC from peak to last milking Milk AUC from peak = $kg \times number$ of days from peak to last milking

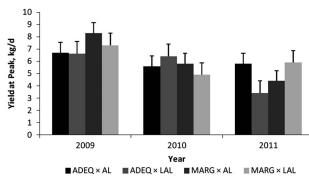


Figure 2 Yield at peak (kg/day; mean \pm SEM) for a *in utero* nutrition (adequate (ADEQ) or marginal (MARG) winter supplementation consisting of alfalfa hay (1.8 or 1.2 kg/day) for 80 days before parturition) \times postweaning heifer development treatment (either *ad libitum* (AL) or 80% of *ad libitum* (less than *ad libitum* (LAL)) feed availability for a 140-day development period) \times year (2009–11) interaction (P = 0.01).

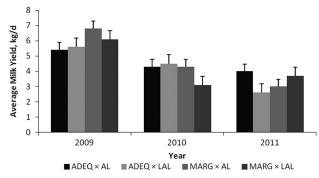


Figure 3 Average milk yield (kg/day; mean \pm SEM) for a *in utero* nutrition (adequate (ADEQ) or marginal (MARG) winter supplementation consisting of alfalfa hay (1.8 or 1.2 kg/day) for 80 days before parturition) \times postweaning heifer development treatment (either *ad libitum* (AL) or 80% of *ad libitum* (less than *ad libitum* (LAL)) feed availability for a 140-day development period) \times year (2009–11) interaction (P=0.01).

the 1st and last year and the highest in the 2nd year (3.4 to 6.6 kg/day). Primiparous cows that received MARG *in utero* nutrition and developed on AL had the highest peak yield in year 1 and linearly declined (8.3 to 4.4, kg/day) in subsequent years. Lastly, primiparous cows that received MARG *in utero* nutrition and developed on LAL had the second highest peak milk yield in the 1st year of the study, the lowest peak yield in year 2 and the highest peak yield the final year of the study (7.3, 4.9 and 5.9 kg/day, respectively).

In utero and post-weaning heifer development milk constituents

No changes in milk urea N were measured for *in utero* nutrition (P = 0.51) or post-weaning heifer development treatment (P = 0.34; Table 2).

Average daily yields of milk constituents (protein, lactose, fat, solids non-fat, g/day) exhibited an *in utero* nutrition \times post-weaning heifer development treatment \times year interaction for peak milk yield ($P \le 0.04$; Figures 3 and 4). As would be expected, milk constituent yields followed similar trends to average daily and peak milk yield. In general, average daily yield and yields of constituents declined

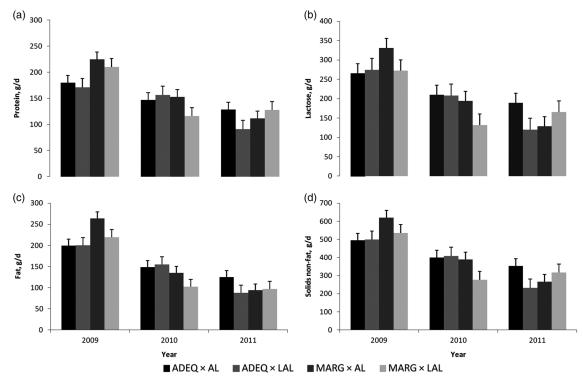


Figure 4 Average milk constituent yield (g/day; mean \pm SEM) for *in utero* nutrition (adequate (ADEQ) or marginal (MARG) winter supplementation consisting of alfalfa hay (1.8 or 1.2 kg/day, respectively) for 80 days before parturition) \times post-weaning heifer development treatment (either *ad libitum* (AL) or 80% of *ad libitum* (less than *ad libitum* (LAL)) feed availability for a 140-day development period) \times year (2009–11) interaction for (a) protein: P = 0.01, (b) lactose: P = 0.01), (c) fat: P = 0.04 and (d) solids non-fat: P = 0.01.

from year 1 to year 3 coincident with the amount of precipitation (Figure 1). As precipitation increased from year 1 to year 3, milk and nutrient constituents' yields decreased. Primiparous cows reared *in utero* on the ADEQ and subsequently receiving post-weaning heifer development treatment of AL were least responsive to environmental changes across years. However, in certain years cows managed in this manner may not produce as much milk and supply as much nutritional constituents. Furthermore, primiparous cows subjected to the ADEQ treatment while they were *in utero* and subsequently to LAL treatment during their post-weaning development may be the more susceptible to changes in environmental conditions.

Appropriate post-weaning pre-pubertal development depends on heifers obtaining an adequate BW before breeding. However, there is great latitude in how this can be achieved (Clanton et al., 1983). Development of heifers at an increased rate of gain post weaning can lead to low pregnancy rates (Ferrell, 1982) and impaired milk production (Hixon et al., 1982; Sejrsen and Foldager, 1992; Sejrsen et al., 2000). However, Endecott et al. (2013) suggests changes in mature BW have occurred over the decades and the recommendation for heifer target BW before breeding of 60% to 65% of mature weight may be different today than when first reported. Recent studies have shown that developing heifers to a lighter targeted BW of 50% to 57% allowed for reductions in heifer development costs and acceptable reproductive performance (Roberts et al., 2009a; Mulliniks et al., 2013; Waterman et al., 2014). The question then

arises; how is first parity lactation impacted by this change in growth curve of heifers developed at BW <60% of mature BW? In the present study, differences in total milk yield were not observed as a consequence of how heifers were reared after weaning or by *in utero* nutritional management during gestation. However, nutritional environment in which heifers were reared (year) was a major contributor to variation in total milk yield. In years, when precipitation was highest during the onset of lactation, milk yield was the lowest. This same response to year was evident in the number of days to reach peak milk yield where peak milk yield occurred earlier in the wettest year.

In conclusion, an *in utero* nutrition effect, implemented during the last 80 days of gestation was not observed for subsequent first lactation milk production in primiparous beef cows. Furthermore, the designed 20% reduction in harvested feed input during post-weaning development (on a common BW basis) ultimately accounted for a 27% decrease in overall feed input to heifers receiving LAL. This measured decrease in feed input for LAL heifers contributed to an earlier peak in milk production. Thus, no detrimental effects in first parity milk yield due to reduction in feed nutrient inputs during *in utero* or post-weaning heifer development were detected.

Acknowledgements

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