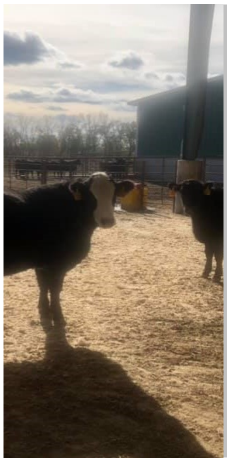

Effect of Rate of Gain During Early Gestation on Colostrum and Milk Composition in Beef Heifers

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The objectives of this study were to evaluate the impact of feeding an energy/protein supplement to replacement heifers to achieve a moderate rate of gain during the first trimester of gestation (84 days) on composition of colostrum and milk, and milk production. Developing heifers to a moderate rate of gain decreased somatic cell count in colostrum and increased the percent of protein in milk; however, no effects were observed on milk production measured via a weigh-suckle-weigh procedure in this study.

Summary

We hypothesized that the rate of gain during the first 84 days of gestation would affect composition of colostrum and milk, and increase milk production in moderate-gain heifers. At breeding, 45 Angus-based heifers received a basal total mixed ration allowing 0.63 pound/day of gain (low gain [LG], n = 23) or a basal diet plus starch-based supplement allowing 1.75 pounds/day of gain (moderate gain [MG], n = 22) for 84 days.

Heifers then were managed on a common diet until parturition. Colostrum samples (50 milliliters [mL]) were collected before first suckling. Milk samples (50 mL) were collected six hours after calf removal on days 62 ± 10 and 103 ± 10 postpartum.

Samples were collected by stripping each teat 15 to 20 times after discarding the first five strips. At day 103, sampling techniques were compared by collecting a second sample after 1 mL oxytocin administration and 90 seconds of lag time.

Data were analyzed using the GLM procedure of SAS. Fat, protein, somatic cell count (SCC), milk urea nitrogen and other solids were analyzed in colostrum for effect of treatment, whereas milk composition was evaluated for effects of treatment, day and their interaction. Heifer was the experimental unit and significance was set at $P \leq 0.05$. Colostrum SCC was greater ($P = 0.05$) in LG (6,949 ± 739 cells × 10^3/mL) than MG (4,776 ± 796 cells × 10^3/mL). In milk, protein and other solids were greater ($P \leq 0.03$) in MG (3.02 ± 0.03 and 6.20 ± 0.02%, respectively) than LG (2.87 ± 0.03 and 6.14 ± 0.02%, respectively).

On day 103, oxytocin administration and extended lag time after teat stimulation (0.96 ± 0.05%) increased fat content in milk ($P < 0.01$) compared with immediate milk sample collection (0.34 ± 0.05%). We conclude that nutrition during early gestation had a sustained impact on milk composition, and techniques of oxytocin administration result in greater milk fat content.

Introduction

In cattle, the development of the mammary gland begins during embryonic development, with the majority of its growth occurring during the last trimester of gestation. By parturition, all components of the gland are established in the fetus, including vascular, lymphatic, connective and adipose tissues (Rowson et al., 2012).

In the heifer dam, the majority of apparent mammary growth occurs during the last trimester of gestation and is completed at parturition (Rowson et al., 2012; Davis, 2017). Therefore, optimal development and growth of the mammary gland during gestation is essential to ensure maximized milk production in future lactations (Meyer et al., 2011).

Additionally, the mammary gland is a key tissue ensuring the transfer of nutrients and immunoglobulins to the neonatal calf (Neville et al., 2010; Geiger, 2020). Because of the importance of a dam's milk production on her calf's weaning weight (Sapkota et al., 2020), optimizing milking potential is crucial.

Milk is produced in the secretory tissue of the alveoli; however, milk's nutritional constituents, and consequently composition, vary depending on place of storage in the udder. In contrast to casein micelles (protein), which are small enough to passive transfer from the alveoli into the cistern, milk fat globules are larger and require active expulsion from the alveoli. Therefore, fat content is greater in the alveoli than in the cistern, whereas protein content is similar across the two storage sites.

Milk letdown is initiated by oxytocin, which is released from the pituitary gland in response to tactile stimulation of the udder and causes the myoepithelial cells around the alveoli to contract and eject the milk stored there into the duct system and cistern (Bruckmaier and Blum, 1998; McKusick et al., 2002; Mačuhová et al., 2004). However, an approximate one- to two-minute lag period occurs between the release of oxytocin and milk expulsion (Bruckmaier and Blum, 1998).

Milk composition can be influenced by multiple factors, with milk fat being the component that can vary the most as a result of environmental and physiological factors, especially nutrition (Bauman and Griinari, 2001).

Fatty acids in milk can stem from two different sources. The short and medium chain fatty acids result from *de novo* synthesis in the mammary gland from carbon sources, including acetate, whereas the longer chain fatty acids originate from preformed circulating fatty acids in the blood (Bauman and Griinari, 2001; Wijesundera et al., 2003). When high-concentrate/low-fiber diets or diets high in plant oils are fed to cattle, ruminal biohydrogenation pathways of poly-unsaturated fatty acid may be shifted from *trans*-11 to *trans*-10 isomers (Fougère et al., 2018; Fougère and Bernard, 2019), with *trans*-10 isomers having an inhibitory effect in milk fat synthesis (Baumgard et al., 2000; Medeiros et al., 2010).

Based on the lack of information in the literature regarding maternal nutrition during early gestation and its effect on lactation, we evaluated the impacts of low and moderate gain during the first 84 days of gestation on composition of colostrum and milk, and milk production.

Experimental Procedures

All animal procedures were approved by the Institutional Animal Care and Use Committee at North Dakota State University.

Forty-five Angus-based heifers (initial body weight [BW] = 818.2 ± 8.7 pounds) were estrus synchronized using a Select Synch plus CIDR protocol and bred via artificial insemination to female-sexed semen from a single sire. At breeding, heifers were blocked by antral follicle count, ranked by BW and assigned to one of two treatments: 1) a basal total mixed ration (TMR; low gain [LG] 0.63 pound/day; n = 23) or 2) the basal TMR diet with the addition of a starch-based energy/protein supplement mixed into the diet (moderate gain [MG] 1.75 pounds/day; n = 25, Table 1).

Heifers were fed individually using the Insentec Feeding System (Hokofarm B.V., Marknesse, The Netherlands). Heifers were weighed on two consecutive days at the beginning and end of the feeding trial, and every 14 days throughout the 84-day period prior to morning feeding, then on days 164, 234, 262, and at the time of calving, pasture turnout and weaning.

Table 1. Dietary ingredients and nutrient composition of the total mixed ration fed to beef heifers during the first 84 days of gestation.

	Treatment	
Item	**LG[1]**	**MG[2]**
Ingredient, % of DM		
Corn silage	37	29
Prairie hay	53	41
DDGS	10	5
Energy/protein supplement	–	25
Chemical composition, %		
Ash	12.57	9.57
Crude protein	10.49	11.57
ADF	36.97	29.38
NDF	61.12	50.68
Fat	1.98	3.48
Calcium	0.95	0.78
Phosphorus	0.40	0.41

[1] Low gain; heifers fed a basal TMR containing a commercially available mineral supplement (Purina® Wind & Rain® Storm® All-Season 7.5 Complete Mineral, Land O'Lakes Inc., Arden Hills, Minn.) fed at a rate of 4 ounces per head per day, targeting gain of 0.63 pound/day.

[2] Moderate gain; heifer fed basal TMR plus an energy/protein supplement formulated with a blend of ground corn, DDGS, wheat midds, fish oil and urea, targeting gain of 1.75 pounds/day.

At calving, a 50-mL colostrum sample was collected from each heifer, before calves suckled for the first time, and placed into a DHIA plastic milk vial. For sample collection, we stripped each teat 15 to 20 times after discarding the first five strips. Colostrum samples were mixed thoroughly to ensure equal distribution of the preservative in the vials throughout the samples, which then were stored at 4 C until further analysis.

At day 62 ± 10 postpartum, we estimated milk production using a 12-hour weigh-suckle-weigh procedure. Briefly, dams and calves were assigned to two groups of 23 and 22 pairs each. At midnight, we separated calves from their dams. At 6 a.m. the next morning, calves were allowed to nurse their dams until satiety (about 30 minutes) to establish similar milking status across the dams.

Then pairs were separated for two six-hour time periods. After each six-hour window, calves were weighed before and immediately after suckling until satiety (about 30 minutes). The difference between the pre- and post-suckling calf weights was recorded as the estimated milk production of the dam for each of the six-hour time periods.

To estimate 24-hour milk production, milk production for the two six-hour separation periods was added together and multiplied by 2 (Shee et al., 2016). Before allowing the calves to suckle their dams at 6 a.m., we collected a 50-mL milk sample into DHIA vials by stripping each teat 15 to 20 times after discarding the first five strips. Samples were mixed thoroughly and stored at 4 C until further analysis.

At day 103 ± 10 postpartum, dams and calves were separated for six hours and a 50-mL milk sample was collected following the same protocol used at day 62 postpartum. Immediately following the collection of the milk sample, we administered oxytocin (1 mL intramuscularly) to each dam and waited for 90 seconds before collecting another 50-mL milk sample to compare sampling protocols.

All samples were shipped to a DHIA milk laboratory (Stearns County DHIA Lab, Sauk Centre, Minn.) within 10 days (colostrum) and five days (milk) after sample collection for analysis of composition of colostrum and milk (fat, protein, somatic cell count [SCC], milk urea nitrogen [MUN] and other solids).

Statistical Analysis

Heifer BW was analyzed as repeated measures in time using the MIXED procedure of SAS for effects of treatment, day and a treatment × day interaction.

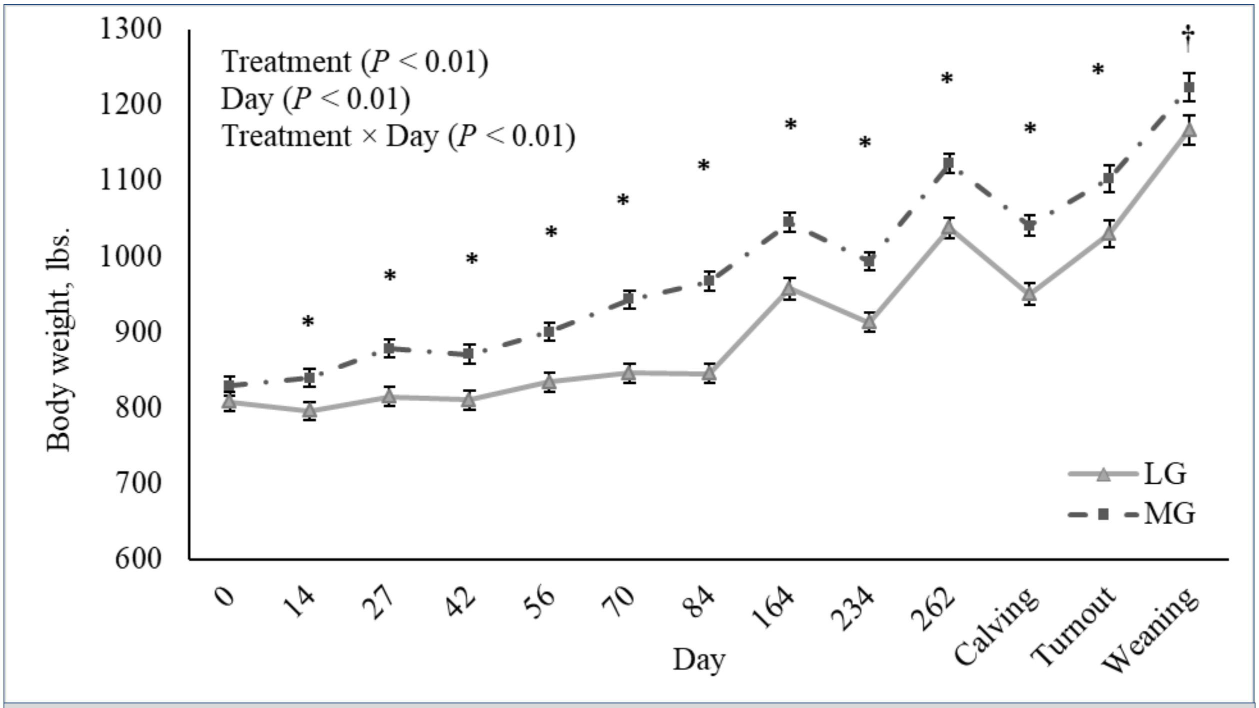

Figure 1. Impact of nutritional treatment on body weight of heifers managed at two rates of gain (low gain [LG], 0.63 pound/day; moderate gain [MG], 1.75 pounds/day) for 84 days, followed by common management for the duration of gestation and lactation. *Within day treatments differ (P < 0.01), † with day treatment differ (P = 0.04).

Colostrum composition and milk production were analyzed using the GLM procedure of SAS. Milk composition at days 62 ± 10 and 103 ± 10 was analyzed using the GLM procedure of SAS for effects of treatment, day, and a treatment × day interaction.

Further, milk composition at day 103 ± 10 was analyzed using the GLM procedure of SAS for effects of treatment, oxytocin and a treatment × oxytocin interaction (SAS Inst. Inc., Cary, N.C.). Heifer was considered the experimental unit in all analyses and significance was set at $P \leq 0.05$.

Results and Discussion

Heifer BW was affected by a treatment × day interaction ($P < 0.01$), being similar at initiation of treatment, diverging by day 14 ($P = 0.01$) and was 122.1 pounds greater for MG heifers at day 84 ($P < 0.01$; Figure 1). Although heifers were managed as a single group beginning at day 85, the weight divergence continued throughout calving until weaning, at which time heifers in the MG treatment remained 90.4 pounds ($P < 0.01$) and 56.1 pounds ($P = 0.04$) heavier than LG heifers at calving and weaning, respectively.

In colostrum (Table 2), we observed an effect of maternal treatment on SCC ($P = 0.05$), which was lower in MG heifers than in LG heifers; however, the percent of fat ($P = 0.11$), protein ($P = 0.40$), other solids ($P = 0.17$) and MUN ($P = 0.29$) were not impacted by rate of gain during the first 84 days of gestation. Somatic cells in colostrum and milk include epithelial cells and leukocytes (macrophages, neutrophils and lymphocytes), with the majority of somatic cells in milk being leukocytes (Kelly et al., 2000).

Consequently, SCC is an indicator of colostrum and milk quality, and a measure of inflammation and infection in the udder. Somatic cell score is greater in colostrum than in milk, which may be caused by cells passing through leaky tight junctions present in the mammary epithelium, which close when milk production increases (Nguyen and Neville, 1998; McGrath et al., 2016).

The SCC in colostrum in this study was far greater than observed by others. For instance, values reported for SCC in beef cattle fed a control diet or a nutrient-restricted diet during the first 82 days of gestation were lower than values reported in the current study (1,276 and 1,043 cells × 10^3/mL for control and restricted, respectively; Noya et al., 2019).

The high SCC values could have been a result of the sampling protocol used, as we did not milk out the entire udder. However, similar to our study, Noya et al. (2019) did not observe any effect of maternal nutritional treatment during early gestation on percent of fat and protein in colostrum, either.

Maternal dietary treatment did not affect milk production on day 62 postpartum ($P = 0.67$;

Table 2. Colostrum composition of beef heifers as influenced by rate of gain (low gain [LG], 0.63 pound/day; moderate gain [MG], 1.75 pounds/day) during the first 84 days of gestation.

	Treatment[1]			
Item	**LG**	**MG**	**SEM[2]**	***P*-value**
Fat, %	5.7	6.7	0.47	0.11
Protein, %	13.6	14.3	0.70	0.40
Somatic cell count, cells × 10^3/mL	6,949	4,776	796	0.05
Milk urea nitrogen, mg/dL	1.7	0.6	0.83	0.29
Other solids, %[3]	4.3	4.5	0.1	0.17

[1] Treatment: low-gain heifers (LG) fed a basal TMR containing a commercially available mineral supplement (Purina® Wind & Rain® Storm® All-Season 7.5 Complete Mineral, Land O'Lakes Inc., Arden Hills, Minn.) fed at a rate of 4 ounces per head per day, targeting gain of 0.63 pound/day; moderate gain-heifers (MG) fed basal TMR plus an energy/protein supplement formulated with a blend of ground corn, DDGS, wheat midds, fish oil and urea, targeting gain of 1.75 pounds/day.

[2] SEM = Standard error of the mean (LG, n = 23; MG, n = 22).

[3] Values for other solids include lactose and ash.

LG: 10.6 ± 0.91 pound/day; MG: 11.2 ± 0.92 pound/day), but influenced milk composition on days 62 and 103 postpartum (Table 3). Moderate-gain heifers had greater percentage of milk protein (*P* < 0.01) and other solids (*P* = 0.03) than LG heifers. Further, the percent of fat and other solids in milk decreased from day 62 to day 103 postpartum (*P* < 0.01), whereas the percent of protein in milk and MUN increased for the same time periods (*P* < 0.01).

Kennedy et al. (2019) used a portable milking machine to determine milk yield and milk composition at day 44 of lactation in beef cows receiving a control diet or the control diet plus dried distillers grains during late gestation. They did not observe differences in milk production or milk composition, but the percent of milk protein was similar to our values (3.08 ± 0.07% for control and 2.98 ± 0.07% for supplement; Kennedy et al., 2019).

At day 103 postpartum, using a sampling technique that included oxytocin administration and an extended lag time of 90 seconds after teat stimulation, we observed an increased percent of milk fat (*P* < 0.01) compared with collecting an immediate sample without oxytocin injection (Table 4). However, oxytocin administration and the extended lag time did not affect the percent of milk protein (*P* = 0.98).

Both observations make sense in regard to the anatomy of the mammary gland and the role that oxytocin plays in the milk ejection process. Regardless of the sampling technique used, milk fat concentrations were extremely low and do not appear representative of the milk fat that calves have access to when compared with results by Kennedy et al. (2019), who reported fat concentrations greater than 4% in beef cows (4.11 ± 0.33% for control and 4.21 ± 0.33% for supplement). Therefore, future sampling techniques should focus on milking at minimum an entire quarter to obtain a better representation of nutrients in milk.

Acknowledgments

The authors express their gratitude for the many personnel involved in this project, including staff at the Central Grasslands Research Extension Center and the Beef Cattle Research Complex, and undergraduate students for assisting with animal handling and data collection.

Table 3. Milk composition of beef heifers at days 62 ± 10 and 103 ± 10 postpartum as influenced by rate of gain (low gain [LG], 0.63 pound/day; moderate gain [MG], 1.75 pounds/day) during the first 84 days of gestation.

	LG[1]		MG[2]			*P*-values		
Item	**d 62[5]**	**d 103[6]**	**d 62**	**d 103**	**SEM[3]**	**Treatment**	**Day**	**Treatment × Day**
Fat, %	0.55	0.35	0.45	0.34	0.044	0.23	<0.01	0.28
Protein, %	2.75	3.0	2.92	3.12	0.045	<0.01	<0.01	0.53
Somatic cell count, cells × 10^3/mL	36.65	88.09	33.59	57.9	28.76	0.55	0.18	0.63
Milk urea nitrogen, mg/dL	4.11	11.15	3.95	10.11	0.425	0.15	<0.01	0.29
Other solids, %[4]	6.20	6.08	6.26	6.13	0.027	0.03	<0.01	0.87

[1] Low gain; heifers fed a basal TMR containing a commercially available mineral supplement (Purina® Wind & Rain® Storm® All-Season 7.5 Complete Mineral, Land O'Lakes Inc., Arden Hills, Minn.) fed at a rate of 4 ounces per head per day, targeting gain of 0.63 pound/day.

[2] Moderate gain; heifers fed basal TMR plus an energy/protein supplement formulated with a blend of ground corn, DDGS, wheat midds, fish oil and urea, targeting gain of 1.75 pounds/day.

[3] SEM = Standard error of the mean (LG, n = 23; MG, n = 22).

[4] Values for other solids include lactose and ash.

[5] Milk sample collected at day 62 ± 10 postpartum.

[6] Milk sample collected at day 103 ± 10 postpartum.

Table 4. Percent of milk fat and protein in beef heifers at day 103 ± 10 postpartum as influenced by sampling technique and rate of gain (low gain [LG], 0.63 pound/day; moderate gain [MG], 1.75 pounds/day) during the first 84 days of gestation

	LG[1]		MG[2]			*P*-values		
Item	Pre-Oxytocin[4]	Post-Oxytocin[5]	Pre-Oxytocin[4]	Post-Oxytocin[5]	SEM[3]	Treatment	Oxytocin	Treatment × Oxytocin
Fat, %	0.35	0.88	0.34	1.03	0.078	0.23	<0.01	0.28
Protein, %	3.00	3.00	3.12	3.12	0.043	<0.01	<0.01	0.53

[1] Low gain; heifers fed a basal TMR containing a commercially available mineral supplement (Purina® Wind & Rain® Storm® All-Season 7.5 Complete Mineral, Land O'Lakes Inc., Arden Hills, Minn.) fed at a rate of 4 ounces per head per day, targeting gain of 0.63 pound/day.

[2] Moderate gain; heifers fed basal TMR plus an energy/protein supplement formulated with a blend of ground corn, DDGS, wheat midds, fish oil and urea, targeting gain of 1.75 pounds/day.

[3] SEM = Standard error of the mean (LG, n = 23; MG, n = 22).

[4] Milk sample collected before injection of 1 mL of oxytocin and a 90 second lag time.

[5] Milk sample collected after administration of 1 mL of oxytocin and a 90 second lag time.

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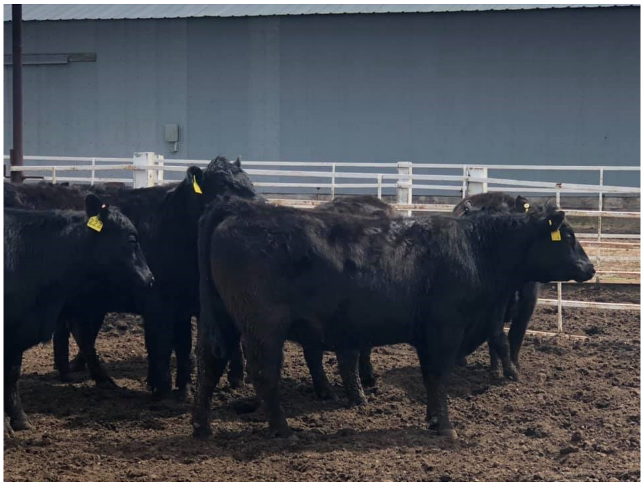

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