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Does β-hydroxybutyrate concentration influence conception date in young postpartum range beef cows?[1]

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ABSTRACT: Cows in negative energy balance after calving often have reduced reproductive performance, which is mediated by metabolic signals. The objective of these studies was to determine the association of serum metabolites, days to first postpartum ovulation, milk production, cow BW change, BCS, and calf performance with conception date in spring-calving 2- and 3-yr-old beef cows grazing native range. In Exp. 1, cows were classified by conception date in a 60-d breeding season as early (EARLY; conceived in first 15 d of breeding) or late conception (LATE; conceived during the last 45 d of breeding). Beginning on d 35 postpartum, blood samples were collected twice per week for serum metabolite analysis and progesterone analysis to estimate days to resumption of estrous cycles. As a chute-side measure of nutrient status and glucose sufficiency, whole-blood β-hydroxybutyrate (BHB) concentrations were measured 14 ± 2 d before breeding. In Exp. 2, cows were classified by subsequent calving date resulting from a 55 ± 2 d breeding season as conceiving either early (EARLY; conceived in first 15 d of breeding) or late (LATE; conceived during the remaining breeding season). Blood samples were collected in 2 periods, 30 ± 4 d before calving and 14 ± 3 d before the initiation of breeding, to determine circulating concentrations of IGF-I and BHB. In Exp. 1, BHB and serum glucose concentrations were less ($P \leq 0.04$) in EARLY cows than LATE cows. Serum insulin concentrations were greater ($P = 0.03$) in EARLY cows relative to LATE cows. Milk production and composition did not differ ($P \geq 0.24$) by conception date groups. In Exp. 2, cow age × sample period × conception date interaction ($P < 0.01$) occurred for serum BHB concentrations. Serum BHB concentrations were similar ($P > 0.10$) for 2-yr-old cows (in greater nutritional plane compared with Exp. 1) regardless of their conception date classification and sampling period. However, precalving serum BHB concentrations were greater ($P < 0.01$) for LATE than EARLY in 3-yr-old cows with no difference ($P = 0.86$) at prebreeding. Serum IGF-1 concentrations were greater ($P < 0.01$) for EARLY cows relative to LATE cows at precalving and prebreeding. This study indicates that blood BHB concentrations during times of metabolic dysfunctions may provide a more sensitive indicator of energy status than body condition, predicting rebreeding competence in young beef cows as measured by interval from calving to conception.

Key words: beef cows, β-hydroxybutyrate, conception date

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INTRODUCTION

Lactating beef cows, especially when grazing primarily dormant range, experience negative energy balance (Mulliniks et al., 2011). Changes in blood metabolites and metabolic hormones during early lactation, resulting from negative energy balance after calving, contribute to endogenous signals to allow or inhibit reproduction (Beam and Butler, 1999). DiCostanzo et al. (1999) demonstrated that intraruminal infusion of acetate for 96 h in ovariectomized heifers experiencing negative energy balance resulted in increased plasma concentrations of acetate, whole-blood β-hydroxybutyrate, and NEFA, which reduced mean concentrations and pulse amplitude of LH. In dairy cows, a decreased serum β-hydroxybutyrate concentration was associated with increased pregnancy rates after first AI (Walsh et al., 2007) and decreased interval to first ovulation (Reist et al., 2000).

Improved efficiency of nutrient utilization or energy metabolism in young cows as identified by faster serum glucose and acetate clearance rates has been reported to increase reproductive performance in beef cows (Waterman et al., 2006; Mulliniks et al., 2011). However, the metabolic signals mediating reproduction is not fully understood. The hypothesis of our research was that young (2- and 3-yr-old) beef cows grazing native dormant range that are very susceptible to metabolic dysfunction as shown by increased concentrations of circulating β-hydroxybutyrate would conceive later in the breeding season because of negative energy balance (from the disconnect involving reduced nutrient intake in comparison with metabolic load of lactation). Therefore, the objective of this study was to determine the association of serum metabolite concentrations, days to first postpartum ovulation, milk production, cow BW and BW change, BCS, and calf growth with conception date in 2- and 3-yr-old beef cows grazing dormant native range.

MATERIALS AND METHODS

All animal handling and experimental procedures were in accordance with guidelines set by the Institutional Animal Care and Use Committee of New Mexico State University (Exp. 1) and the ARS-USDA Fort Keogh Livestock and Range Research Laboratory (**LARRL**) Animal Care and Use Committee (Exp. 2).

Study Site

Experiment 1. The experiment was conducted over 3 yr at the Corona Range and Livestock Research Center (**CRLRC**) of New Mexico State University. Average elevation for CRLRC is 1,900 m with average rainfall of 370 mm, of which 50% occurs in July and August (Torell et al., 2008). Forages at this study site were primarily warm-season grasses and consisted of blue grama (*Bouteloua gracilis*), three-awns (*Aristida* spp.), and common wolf tail (*Lycurus phleoides*). The pasture was 762 ha and contained approximately 355 kg/ha of standing forage (A. Cibils, New Mexico State University, personal communication). All pastures were stocked at a rate that was 50% less than the Natural Resources Conservation Service (**NRCS**) recommended rate so that forage availability was assumed not to limit cow productivity (NRCS-USDA, 2002). Three ruminally cannulated cows were used to collect diet extrusa samples for analysis of CP (AOAC, 2000) and NDF (Van Soest et al., 1991). Diet extrusa samples were collected in April before breeding via ruminal evacuation techniques described by Lesperance et al. (1960). Extrusa samples averaged (OM basis) 11.3%, 5.1%, and 8.1% CP and 80.0%, 78.6%, and 85.9% NDF for yr 1, 2, and 3, respectively.

Experiment 2. The experiment was conducted at LARRL. Experiment 2 was designed to determine if the results from Exp. 1 could be replicated in a different environment and management with similar calving and breeding dates. The experiment was conducted at the LARRL at an average elevation of 730 m. Native vegetation on the 22,500-ha research laboratory is primarily cool-season grasses consisting of a needlegrass-wheatgrass (*Stipa-Pascopyrum*) mix. Long-term average precipitation is 343 mm with about 60% to 70% occurring during the mid-April through mid-June growing season. Average annual forage standing crop at the study site is 870 ± 14 kg/ha (Grings et al., 2005).

Animals

Experiment 1. Cows were 2- and 3-yr-old ($n = 131$), primarily of Angus breeding with some Hereford influence. Four cows were not pregnant at the end of the experiment and therefore were not used in the analysis. Management before calving and after calving was similar in all years and among all cows. The 2- and 3-yr-old cows were managed together as 1 herd, but separately from the mature cow herd. At least 60 d before the initiation of calving, cows were fed 1.6 kg/cow of a 36% CP cube once per week with no additional harvested feedstuffs. Cow-calf pairs were moved to a common pasture within 10 d after calving, where a 36% CP supplement was fed 2 times/wk at a rate of 908 g/(cow·d). Postpartum supplementation was provided for 70 ± 3 d across the 3-yr study. In mid-May of each year, estrus was synchronized using a controlled internal drug-releasing (**CIDR**) device (Eazi-Breed CIDR, Pfizer Animal Health, New York, NY) with the CO-Synch protocol. Cows were administered a single 2-mL intramuscular (**i.m.**) injection of

GnRH (Cystorelin, Merial, Iselin, NJ), and a CIDR was inserted. After 7 d, the CIDR was removed, and all cows received a single 5-mL i.m. injection of PGF (Lutalyse, Pfizer Animal Health). Approximately 66 h after CIDR removal, all cows were artificially inseminated and were administered a single 2-mL i.m. injection of GnRH (Cystorelin, Merial). A 60-d breeding season was initiated the day after AI in all years, with a bull-to-cow ratio of 1:26.

Experiment 2. Cows used in this experiment were 2- and 3-yr-old (n = 381) from a stable composite population (**CGC**; 1/2 Red Angus, 1/4 Charolais, 1/4 Tarentaise). Two-year-old cows were managed separately from older cows during the winter before calving to allow for a better management of nutritional environment, whereas the 3-yr-old cows were managed with the mature cow herd. For the majority of the winters in this study, forage was readily available for grazing, and the only additional harvested feed provided to the herd was alfalfa cake or hay, depending on year, as a supplemental source of protein. This supplement was fed either daily or every other day to achieve an average of about 1.5 kg/d. During days when access to grazing was limited because of snow cover, cows were fed at a rate equivalent to 9 kg alfalfa hay/d for each cow. In 3 of 4 yr of the study, cows were subjected to a 7-d Co-Synch protocol as described in Exp. 1 to facilitate breeding by AI followed by natural mating for the remaining duration of a 55 ± 2 d breeding season. In yr 4, cows were injected with $PGF_{2\alpha}$ (Lutalyse, Pfizer Animal Health) to induce luteal regression and were artificially inseminated as estrous was detected. Clean-up bulls were used in yr 4 for 56 d after all cows were artificially inseminated.

Experimental Groups

In Exp. 1 and 2, date of conception for the study year was calculated from the calving date of the subsequent year minus 285 d for gestation. Cows were retrospectively classified as early (**EARLY**; calculated to have conceived within the first 15 d of the breeding season, which included both AI and natural breeding) or late conception (**LATE**; calculated to have conceived in the last 45 d of the breeding season in Exp. 1 and the last 41 ± 3 d in Exp. 2). Day 15 was chosen as the division to average an 80-d (postpartum) conception date and give a calving interval of 365 d for EARLY cows. Breeding season in all years started on day of AI.

Sampling and Analyses

Experiment 1. Blood samples (9 mL) were collected twice weekly on days of supplementation (Monday and Friday) via coccygeal venipuncture (Corvac, Sherwood Medical, St. Louis, MO) beginning approximately 35 d postpartum for analysis of progesterone to determine days to first postpartum ovulation (2 or more consecutive progesterone concentrations ≥1.0 ng/mL). Blood samples were collected, cooled, and subsequently centrifuged at 2,000 × g at 4°C for 20 min. Serum was harvested and stored at −20°C in plastic vials for later analysis. Serum was analyzed for progesterone concentration by solid phase RIA (Coat-A-Count, Diagnostic Products Corp., Los Angeles, CA) as described by Schneider and Hallford (1996). Inter- and intra-assay CV were less than 10%.

Serum samples were also analyzed for insulin, glucose, NEFA, IGF-I, and urea N (**SUN**). To evaluate the nutrient status of each cow, serum samples were composited by cow for a prebreeding serum sample. Composited serum samples were analyzed using commercial kits for NEFA (Wako Chemicals, Richmond, VA) and SUN (Thermo Electron Corp., Waltham, MA). Glucose was analyzed with a commercial kit (enzymatic endpoint, Thermo Electron Corp., Waltham, MA). Insulin was analyzed by solid-phase RIA (Count-A-Coat, Siemens Medical Solutions Diagnostics, Los Angeles, CA) as reported by Reimers et al. (1982). Serum IGF-I samples were quantified by double-antibody RIA (Berrie et al., 1995). Inter- and intra-assay CV were less than 10%. As a chute-side measure of nutrient status and glucose sufficiency, whole-blood β-hydroxybutyrate (**BHB**) concentrations were measured (MediSense, Precision Xtra, Abbott Laboratories, Abbott Park, IL; validated by Byrne et al., 2000; Endecott et al., 2004; Voyvoda and Erdogan, 2010) in early May, 2 wk before breeding.

A subsample of cows randomly selected to be an equal representation of age were milked by a portable machine (Porta-Milker, Coburn Company Inc., Whitewater, WI) approximately 57 d postpartum. Milking procedures were a modified weigh-suckle-weigh technique described by Waterman et al. (2006). Milk weights were recorded to calculate 24-h milk production. Milk samples were analyzed for lactose, butterfat, solids nonfat, and protein by Pioneer Dairy Labs (DHIA, Artesia, NM).

Experiment 2. Blood samples (9 mL) were collected via coccygeal venipuncture (Corvac, Sherwood Medical, St. Louis, MO) at 2 time periods: 30 ± 1 d before the start of calving and 14 ± 1 d before start of breeding. Blood handling and storage was similar to that described in Exp. 1. Serum samples were analyzed for IGF-1 and BHB. Serum IGF-I samples were quantified by double-antibody RIA after acid-ethanol extraction of IGF-binding proteins (Funston et al., 1995; Roberts, 2008) using AFP4892898 as the primary antibody and I^{125}-labeled recombinant human IGF-1 purchased from PerkinElmer (Billerica, MA). Inter- and intra-assay CV for IGF-I were 12% and 15%, respectively. As a measure of nutrient status and glucose sufficiency, BHB concentrations were

Table 1. Prebreeding blood ketones, serum metabolites, and milk production for 2- and 3-yr-old cows classified as early or late conception in Exp. 1

Prebreeding measurement	Conception[1]		SEM	P-value
	EARLY	LATE		
n	65	62	—	—
Blood ketones, µmol/L				
Whole-blood β-hydroxybutyrate	297	371	20	0.04
Serum metabolites				
Glucose, mg/dL	52.9	58.1	1.7	0.02
Insulin, ng/mL	0.81	0.56	0.07	0.03
NEFA, mmol/L	501	491	19	0.69
Serum urea N, mg/100 mL	9.07	9.74	0.48	0.32
IGF-I, ng/mL	45.78	42.04	3.46	0.42
Milk, g/d				
24-h milk production	5991	6510	335	0.28
Lactose	298	320	16	0.34
Protein	159	175	9	0.24
Solids-non-fat	512	553	28	0.31
Butterfat	197	215	15	0.41

[1]Conception date was estimated from calving date in the subsequent year: EARLY = conceived within first 15 d of breeding; LATE = conceived during the remainder of the breeding season.

Table 2. Calving date and reproductive measurements for 2- and 3-yr-old cows classified as early or late conception in Exp. 1

Measurement	Conception[1]		SEM	P-value
	EARLY	LATE		
Calving date,[2] Julian d	62	65	2	0.19
Reproductive measurements				
Resumption of estrus,[3] d	71	78	2	0.04
Conception date,[4] d	80	99	1	<0.01
Estrus to conception,[5] d	9	21	2	<0.01
BW nadir, d	59	57	4	0.71
Nadir to estrus,[6] d	12	21	5	0.18

[1]Conception date was estimated from calving date in the subsequent year: EARLY = conceived within the first 15 d of breeding; LATE = conceived during the remainder of the breeding season.

[2]Calving date of the study year.

[3]Interval from calving to resumption of estrus.

[4]Interval from calving to estimated day of conception.

[5]Interval from resumption of estrus to estimated day of conception.

[6]Interval from BW nadir to resumption of estrus.

measured using a commercially available diabetic monitor system as described in Exp. 1.

Animal Performance

In Exp. 1, cows were weighed weekly after calving until termination of the breeding season and at weaning. Days to BW nadir was calculated from the lowest BW after calving. Body condition scores (1 = emaciated, 9 = obese; Wagner et al., 1988) were assigned to each cow by visual observation and palpation at initiation of the study, branding, and weaning by 2 trained technicians. In Exp. 2, cows were weighed, and BCS were assigned to each cow by visual observation and palpation by 2 trained technicians at 2 time periods in relation to calving or breeding: 30 ± 1 d before start of calving and 14 ± 1 d before start of breeding. Calves were weighed at birth, branding, and weaning in each year. Weights at branding and weaning were adjusted to 55- and 205-d age constant BW. Cows were diagnosed pregnant by rectal palpation at weaning or a few weeks later.

Statistical Analysis

Normality of data distribution and equality of variances of measurements were evaluated using PROC UNIVARIATE and the Levene test, respectively. Data were analyzed as a completely randomized design with cow as the experimental unit using the Kenward-Roger degrees of freedom method. The MIXED procedure (SAS Inst. Inc., Cary, NC) was used to test a model that included fixed effects of conception date group, cow age, year, sex of calf, and their interactions. Separation of least squares means was performed by the PDIFF option of SAS when a significant ($P \leq 0.05$) effect of conception date classification was detected.

RESULTS

Experiment 1. At 2 wk before breeding, cows classified as EARLY had less ($P = 0.04$; Table 1) concentrations of BHB than cows classified as LATE. Serum glucose concentrations were decreased ($P = 0.02$) in EARLY cows. Serum insulin concentrations were greater ($P = 0.03$) in EARLY cows, which may have facilitated glucose metabolism. Serum NEFA, SUN, and IGF-1 concentrations were not different ($P > 0.28$) between EARLY and LATE cows.

Twenty-four-hour milk production did not differ ($P = 0.28$; Table 1) between EARLY and LATE cows. Concentrations of milk butterfat, protein, lactose, and solids nonfat also were not different ($P \geq 0.24$) between conception classification groups.

Calving date during the initial year of the study was not different ($P = 0.19$; Table 2) between EARLY and LATE cows. Circulating concentrations of progesterone indicated that interval from calving to first postpartum ovulation was decreased ($P = 0.04$) in EARLY cows. Early conception cows returned to estrus 7 d before LATE cows. In addition, the interval from first postpartum ovulation to conception date was decreased ($P < 0.01$) by 12 d for EARLY cows. Therefore, conception occurred 19 d sooner in the breeding season for EARLY cows ($P < 0.01$).

Table 3. Cow BW and BW change, BCS, and calf BW for 2- and 3-yr-old cows classified as early or late conception in Exp. 1

	Conception[1]			
Measurement	EARLY	LATE	SEM	*P*-value
BCS[2]				
Calving	4.5	4.6	0.04	0.81
Branding	4.0	4.1	0.05	0.12
Weaning	4.6	4.7	0.06	0.80
Cow BW, kg				
Calving	449	454	5	0.41
Nadir	368	368	4	0.97
Begin of breeding	393	388	5	0.39
End of breeding	400	408	5	0.19
Weaning	447	453	5	0.37
Cow BW change, kg				
Calving to begin of breeding	−56	−67	3	0.01
Calving to nadir	−81	−87	4	0.24
Calving to end of breeding	−49	−46	4	0.59
Nadir to end of breeding	32	40	3	0.04
Nadir to weaning	79	85	3	0.14
Calving to weaning	−2	−1	4	0.87
Calf BW, kg				
Birth	34	33	1	0.46
Branding[3]	70	68	2	0.49
Weaning[4]	203	205	4	0.67

[1]Conception date was estimated from the subsequent years calving date: EARLY = conceived within first 15 d of breeding; LATE = conceived during the remainder of the breeding season.

[2]For BCS, 1 = emaciated, 9 = obese; Wagner et al. (1988).

[3]Calf BW at branding, adjusted for 55-d BW.

[4]Calf BW at weaning, adjusted for 205-d BW.

Table 4. Cow BW, BCS, calf BW, and circulating concentrations of IGF-I for 2- and 3-yr-old cows classified as early or late conception in Exp. 2

	Conception date[1]			
Measurement	EARLY	LATE	SEM	*P*-value
n	145	160	—	—
Calving date, Julian d	90	92	5	0.49
Conception date[2], d	81	111	5	<0.001
Cow BW, kg				
Precalving	451	448	7	0.80
Prebreeding	426	426	7	0.97
Cow BCS[3]				
Precalving	5.0	5.0	0.1	0.83
Prebreeding	4.5	4.4	0.1	0.43
Calf BW, kg				
Birth	33	34	1	0.25
Weaning[4]	198	224	3	<0.01
IGF-I, ng/mL	144	130	5	<0.01

[1]Conception date was estimated from calving date in the subsequent year: EARLY = conceived within the first 15 d of breeding; LATE = conceived during the remainder of the breeding season.

[2]Interval from calving to estimated conception.

[3]1 = emaciated, 9 = obese; Wagner et al. (1988)

[4]Weaning BW adjusted for 205-d BW.

Days to BW nadir and interval from BW nadir to first postpartum ovulation were not influenced ($P \geq 0.18$) by conception classification groups. Cow BW and BCS were similar between EARLY and LATE cows at all measurement times ($P \geq 0.12$; Table 3). However, BW loss from the initiation of the study to the beginning of breeding was greater ($P = 0.01$) in LATE cows, suggesting differences in negative energy balance due to associated metabolic imbalances. Subsequently, LATE cows did gain more ($P = 0.04$) from BW nadir to the end of breeding, with no differences ($P > 0.14$) in BW change at any other time period. Calf BW at birth, branding, and weaning were not influenced ($P \geq 0.46$) by conception classification group.

Experiment 2. Calving date during the initial year of the study was not different ($P = 0.49$; Table 4) between EARLY and LATE cows. Conception date for EARLY cows were 30 d earlier ($P < 0.01$) than LATE cows. This difference is expected due to classification of conception date groups.

Body condition score and cow BW were not different ($P \geq 0.43$; Table 4) at calving and breeding between EARLY and LATE cows. Calf BW at birth was not different ($P = 0.25$) for EARLY and LATE cows. However, 205-d adjusted BW of calf at weaning was greater ($P < 0.01$) for LATE cows, indicating a potential for differences in milk production.

Circulating concentrations of IGF-I were greater ($P < 0.01$; Table 4) in EARLY cows. A cow age × conception date × sample time interaction ($P < 0.01$; Table 5) occurred for serum BHB concentrations. In 2-yr-old cows, serum BHB concentrations did not differ ($P > 0.10$) due to conception date classification or sampling time. However in 3-yr-old cows, precalving serum concentrations of BHB were greater ($P = 0.05$) in LATE rather than EARLY cows.

DISCUSSION

Circulating concentrations of BHB will increase when rate of acetate oxidation is slower than incoming supply of acetate. A condition that can contribute to reduced oxidation of acetate is an inadequate supply of cellular oxaloacetate derived from serum glucose (Kaneko, 1989). Elevated concentrations of BHB are indicative of metabolic dysfunction resulting from poor adaptation to negative energy balance (Herdt, 2000). In dairy cows, decreased serum BHB concentration prebreeding was associated with increased pregnancy rates from first-service AI (Walsh et al., 2007) and decreased interval to first ovulation (Reist et al., 2000). Intraruminal infusion of acetate in ovariectomized heifers expe-

Table 5. Concentrations of β-hydroxybutyrate (BHB) in precalving and prebreeding serum samples from 2- and 3-yr-old cows classified as early or late conception in Exp. 2

	Conception[1]		
Measurement	EARLY	LATE	SEM
Precalving BHB			
2-yr-olds	567[ax]	518[ax]	24
3-yr-olds	595[ax]	710[by]	36
Prebreeding BHB			
2-yr-olds	382[ax]	379[ax]	18
3-yr-olds	387[ax]	363[ax]	28

[a,b]For each interaction within timing of sample, means in rows with different superscripts differ ($P < 0.05$).

[x,y]For each interaction within timing of sample, means in columns with different superscripts differ ($P < 0.05$).

[1]Conception date was estimated from calving date in the subsequent year: EARLY = conceived within the first 15 d of breeding; LATE = conceived during the remainder of the breeding season.

riencing negative energy balance resulted in increased circulating concentrations of acetate, BHB, and NEFA, which reduced mean concentrations and pulse amplitude of LH (DiCostanzo et al., 1999). In agreement, Iwata et al. (2011) reported increased BHB concentrations suppressed LH pulses and proposed that ketone bodies might function as a negative energy signal to inhibit gonadal function through suppression of gonadotropin secretion. Prebreeding BHB concentrations in Exp. 1 were reduced in early conception cows compared with late conception, indicating that BHB concentration may have delayed conception date. However, it is important to note that the concentrations of BHB were 24% to 30% of the values used to diagnosis clinical ketosis (1,200 μmol/L). In Exp. 2, BHB concentrations were not different between early and late conception groups for the 2-yr-old cow herd (managed separately from the cow herd) at precalving and prebreeding sample, which indicates there were other mechanisms that delayed conception not investigated in this study. However, BHB concentrations were greater for late conception 3-yr-old cows before calving compared with early conception 3-yr-old cows. Precalving BHB concentrations were not different between early conception 2- and 3-yr-old cows and late conception 2-yr-old cows, suggesting these cows were in similar nutrient balances. This result is not surprising because these cows experienced reduced nutrient intake precalving, during consumption of dormant forages, and the 2-yr-old cows were managed in a less stressful environment. Overall, results from prebreeding in Exp. 1 and 3-yr-old cows at precalving Exp. 2 indicate that increased BHB in range beef cows may be indicative of their capacity for timely rebreeding, even in different environments and with different management practices.

Glucose has been suggested as one of the most important metabolic substrates required for proper function of reproductive processes in beef cows (Short and Adams, 1988). Richards et al. (1989) suggested that a reduction in plasma glucose concentration may cause inadequate serum concentrations of LH required for normal cyclic ovarian function. In Exp. 1, circulating concentrations of glucose were decreased in early conception cows. This difference in serum glucose concentrations was unexpected because glucose is tightly regulated (Kaneko, 1989). The reduced serum glucose and BHB concentrations suggest that serum glucose tissue clearance rate was faster, and increased tissue uptake allowed for the rate of acetate oxidation to be in better equilibrium with glucose supply, thus preventing a subsequent increase in BHB concentration. However, early conception cows also had increased circulating insulin concentrations, which likely influenced and altered the site of glucose accumulation being either blood or tissue. In addition, increased serum insulin concentrations may have had a positive effect on the restoration of LH pulse frequency (Chagas, 2003).

Circulating NEFA concentrations were not different between early and late conception cows in Exp. 1. In contrast to the current study, Ospina et al. (2010) reported that NEFA concentrations have a stronger association with reproductive performance than BHB in dairy cows. The severity of negative energy balance and diet quality between a dairy cow and range beef cow, due to a 4- to 5-fold greater milk yield, may explain some of these differences. However, Pushpakumara et al. (2003) reported that serum BHB concentration before breeding was a better indicator of metabolic status than serum NEFA and glucose concentrations in dairy cows.

Insulin-like growth factor-I has been suggested to be a better indicator of rebreeding performance of first calf heifers than BCS or BW change (Roberts, 2008). In dairy cows, IGF-I may act as a signal for poor metabolic status to the ovary and reproductive tracts (Pushpakumara et al., 2003). In Exp. 1, IGF-I was not different between EARLY and LATE cows; however, IGF-I was greater for EARLY cows in Exp. 2. The contrasting results in experiments in this study may be due to timing of sample collections and environmental and management differences associated with seasonality of rainfall and its effect on seasonal forage growth and quality and combined or separate herds of 2- and 3-yr-old cows. Pushpakumara et al. (2003) reported a tendency for reduced IGF-I concentrations before breeding in cows conceiving later in the breeding season relative to cows conceiving early in the breeding season. Furthermore, Roberts et al. (1997) reported a relationship between IGF-I concentrations and the time of resumption of ovarian cyclicity in postpartum beef cows.

Increased acetate use as inferred by decreased BHB and glucose concentrations indicates differences in whole-blood nutrient concentrations, which could result in decreased precursors required for milk production. In Exp. 1, milk production was not different between conception date groups. Likewise, Pushpakumara et al. (2003) reported no difference in 24-h milk production between dairy cows that become pregnant to an early or late service. However, in Exp. 2, calf BW at weaning was less for EARLY cows than LATE cows, possibly a reflection of differences in milk production between EARLY and LATE cows reflecting divergence in whole-blood metabolite/nutrient dilution.

Calving date of the study year was not different in either experiment for EARLY and LATE cows. However, estimated conception dates were 19 and 30 d earlier for EARLY cows than LATE cows in Exp. 1 and 2, respectively. In Exp. 1, the interval from calving to first postpartum ovulation and interval from estrus to conception were 7 and 12 d earlier for EARLY than LATE cows, respectively. Therefore, increasing the number of young cows becoming pregnant early in the breeding season would be expected to increase their retention rate in the herd by calving early in the subsequent calving seasons with overall improvements in herd longevity.

In both experiments, cow BW and BCS before calving and breeding were not different between EARLY and LATE cows. In Exp. 1, EARLY cows did lose less BW from calving to breeding compared with LATE cows. However, this difference in BW loss and BCS change may be due to differences in efficiency of energy use and adaptation to negative energy balance during early lactation. Body weight and BCS change are considered functional indicators of energy status and reproductive performance after calving (Randel, 1990). Prepartum body energy reserves can be influential in the number of days required to resume estrus and subsequent pregnancy rates in young cows (Spitzer et al., 1995). The results of the present study are in agreement with previous work where no association was observed between BCS at calving and reproductive performance in young range cows when evaluated within a nutritional environment rather than across nutritional treatments (Mulliniks et al., 2012). In the current study, neither BCS nor BW alone influenced reproductive performance. However, BHB concentrations were indicative of reproductive performance and subsequent calving date in both locations with different management and environmental stress when metabolic dysfunctions were present (i.e., increased BHB concentrations). Further research is needed in identifying threshold concentrations of BHB and timing of measurement in young cows for assessing cows that need additional management input (e.g., feed, estrus induction).

Some inconsistencies are evident when comparing age of cow and timing of BHB sample between both experiments. Nonetheless, it would appear that a relationship does exist between BHB concentration and conception date. Results from Exp. 1 and 2 indicate that BHB concentrations may assist cow-calf producers with segregating young cows into early or late conception date groupings. In a review, Wathes et al. (2007) suggested 3 reasons for inconsistent results using blood metabolites as indicators of reproductive efficiency: 1) insufficient numbers of cows, 2) sampling too late after calving, and 3) mixing of data from different age groups. One explanation for this discrepancy in the current study may be due to differences in season of precipitation and type of predominant forage (cool-season vs. warm-season forages) at each research site where Exp. 1 and 2 were conducted. In Exp. 1, the majority of effective precipitation at CRLRC occurs in July and August after breeding is concluded, whereas in Exp. 2, the majority of effective precipitation at LARRL occurs in mid-April through mid-June at the onset of breeding. This difference in timing of precipitation and moisture-stimulated forage growth (quality) is reflected in BW change differences in the studies. Thus, timing of precipitation and forage quality may alter the adaptive mechanisms to negative energy balance (from the metabolic load of lactation). Cow BW at calving were similar between experiments; however, cows in Exp. 1 lost 31 to 44 kg more BW from calving to breeding. In addition, the magnitude of difference in days to conception comparing EARLY to LATE cows between Exp. 1 and 2 was 19 and 30 d, respectively. Therefore, less variation in conception date in EARLY and LATE cows used in Exp. 1 may indicate dissimilarities in fertility, management, or environmental interactions between herds used in each experiment. In Exp. 1, 2- and 3-yr-old cows were managed together in 1 herd, and cows were managed separately in Exp. 2. Although inconsistencies were apparent, BHB concentration was consistently reduced in EARLY cows in both ages in Exp. 1 and with 3-yr-old cows in Exp. 2. It was interesting, however, that cows in Exp. 1 grazing a lower quality of forage with lower calving BCS and increased BW loss after calving had decreased or equal prebreeding BHB concentrations and a narrower conception date interval between treatment groups than in Exp. 2.

Detecting serum metabolite differences during precalving and early lactation was an effective means to segregate cows based on classification of young beef cows according to their conception date. This study indicates that increased BHB concentrations due to metabolic imbalances may be related to or may have a detrimental effect on the interval of resumption of estrus in young beef cows and thereby prolong time until conception. A reduced concentration of BHB was associated with an earlier conception date in 2- and 3-yr-old lactating beef cows in Exp. 1 and 3-yr-old lactating beef cows in Exp. 2. Therefore, BHB concentrations may be a useful indicator of days to resumption of estrus and conception date when metabolic dysfunctions are present. In addition, chute-side measurement BHB may provide producers the opportunity to proactively manage cows (depending on BHB concentration) to improve overall reproductive efficiency.

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