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Feeding Lactose to Increase Ruminal Butyrate and the Metabolic Status of Transition Dairy Cows[1]

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ABSTRACT

Twenty-four multiparous Holstein cows (775 ± 24 kg body weight; 3.4 ± 0.11 body condition score) were used in a randomized complete block design experiment to determine the impact of increased ruminal butyrate from the fermentation of lactose on metabolism and lactation. Dietary treatments were either a corn-based control diet (CON) or a diet containing lactose at 15.7% of diet dry matter (LAC). Experimental diets were fed from 21 d before expected calving through 21 d in milk (DIM). Blood was sampled at –21, –14, –7, –2, 2, 7, 14, and 21 DIM, rumen fluid at –21, –7, and 7 DIM, and liver tissue via biopsy at 7 and 14 DIM. Pre- and postpartum dry matter intake (DMI) through 28 DIM averaged 12.8 and 17.7 kg/d, respectively, and did not differ between treatments; however, cows fed LAC did not exhibit a prepartum decrease in DMI. Milk yield was unaffected by treatments and averaged 45.7 kg/d during the first 70 DIM. Plasma glucose, insulin, and nonesterified fatty acids were not affected by dietary treatments. Feeding LAC increased the ruminal proportion of butyrate both pre- (11.3 vs. 9.2 ± 0.45%) and postpartum (13.0 vs. 10.3 ± 0.67%). Likewise, circulating plasma β-hydroxybutyrate was increased both pre- (6.1 vs. 4.2 ± 0.31 mg/dL) and postpartum (14.6 vs. 8.34 ± 1.7 mg/dL) when feeding LAC compared with CON. Liver lipid content was decreased (8.6. vs. 14.7 ± 1.5% of wet weight) in cows fed LAC relative to those fed CON, whereas liver glycogen was not affected by dietary treatments. Feeding lactose to transition dairy cows increased the proportion of butyrate in the rumen and β-hydroxybutyrate in plasma and decreased liver lipid but did not affect lactation performance.

Key words: lactose, butyrate, β-hydroxybutyrate, transition dairy cow

INTRODUCTION

Research on the feeding and management of transition dairy cows has focused on dietary manipulations to increase ruminal propionate as a means to maximize precursors for hepatic gluconeogenesis. These research efforts have included feeding propionate-based supplements (Mandebvu et al., 2003), decreasing the forage-to-concentrate ratio (Holcomb et al., 2001), and administering drenches containing gluconeogenic substrate (Pickett et al., 2003). Reports delineating effects of manipulation of other ruminal VFA during the periparturient period are limited.

Butyrate is a ketogenic VFA as it is metabolized to BHBA during absorption across the rumen epithelium (Weigand et al., 1975). Other studies have validated this effect on ketone bodies under varying conditions. Andersson and Lundström (1985) found a positive correlation between butyric acid intake from silage high in butyric acid and milk ketone bodies. Likewise, Krehbiel et al. (1992) infused butyrate into the rumen and observed increases in plasma BHBA and decreases in plasma glucose concentrations. Lactose has been shown to ferment to butyrate in the rumen (Schingoethe, 1976). Recently, feeding lactose to lactating dairy cows at 0, 7, and 14% of diet DM increased the molar proportion of butyrate in the rumen (13.9, 16.3, and 18.0, respectively) without placing cows at risk for ketosis as evidenced by changes in plasma BHBA and glucose concentrations (DeFrain et al., 2004).

The relationship between ruminal butyrate and glucose is largely unexplored. β-Hydroxybutyrate has been demonstrated to increase hepatic gluconeogenic activity through its metabolism to acetyl-CoA, an allosteric activator of pyruvate carboxylase (Utter and Keech, 1963). In support of these findings, Black et al. (1966) showed that increased concentrations of ruminal butyrate metabolized by ruminal epithelium to circulating BHBA would serve to spare pyruvate, a glucogenic precursor, from oxidation and enhance net conversion of pyruvate to oxaloacetate. Later, tracer work by Anand and Black (1970) showed that intravenous butyrate injections stimulated gluconeogenesis in cattle. Therefore, we have hypothesized that ruminally produced butyrate could be beneficial to the transition cow by 1)

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sparing the hepatic oxidation of pyruvate and enhancing its conversion to oxaloacetate, 2) sparing glucose use by extramammary tissues (Holtenius and Holtenius, 1996), and 3) providing precursors for fatty acid synthesis (Palmquist et al., 1969). The impact of diets fermenting to butyrate in the rumen of transition dairy cows, however, has not been explored. The objective of this experiment was to determine the impact of increased ruminal butyrate on key indicators of energy metabolism and lactation performance.

MATERIALS AND METHODS

Experimental Design and Feeding and Management of Cows

The experiment was conducted from September 2003 through July 2004 at the South Dakota State University Dairy Teaching and Research Facility (Brookings, SD). Animal care and use was according to a protocol approved by the South Dakota State University Institutional Animal Care and Use Committee. Twenty-four multiparous Holstein cows were used to examine the effects of feeding lactose on DMI, milk production and composition, blood metabolites, and liver chemical composition. Treatments were arranged as a randomized block design and blocked by expected calving date.

At 28 d before expected calving date, cows were assigned to their respective treatment diet (Table 1), either control (**CON**) or lactose (**LAC**; First District Ag Service, Litchfield, MN) at 15.7% of diet DM. The composition of the LAC diet was selected based on prior research (DeFrain et al., 2004) where lactose was included at 14.2% of the diet DM in place of corn and extruded soybeans, and successfully increased ruminal butyrate proportions from 13.9 to 18.0%. To balance diet RUP, dried distillers grains were incorporated into LAC during the first 21 DIM. Cows were adapted to the Calan Broadbent feeding system (American Calan, Inc., Northwood, NH) for 1 wk before initiation of treatments. Experimental diets were fed from 21 d (SD = 5.2) before calving through 21 DIM. From 22 to 70 DIM, all cows were fed a common herd diet. Individual feed intakes were monitored through 28 DIM.

Cows were housed on a wheat-straw bedded pack prepartum and moved to box stalls immediately before calving. After parturition, calves were immediately removed from the dam. Cows remained in box stalls with feed intake recorded daily until their milk was analyzed to be free of antibiotics, at approximately 4 DIM, at which time they were moved to a free-stall barn equipped with the Calan Broadbent feeding system.

Measurements and Collection of Samples

Diets were mixed and fed daily at 0600 h for ad libitum consumption as a TMR. Feed orts were measured, recorded, and discarded before feeding each day, and amounts fed were adjusted to ensure a 10% refusal. Body weight and BCS (1 to 5 in 0.25 increments; Wildman et al., 1982) were recorded on 2 consecutive days, 4 h after feeding on d 22 and 21 before expected calving date, at parturition and 1 DIM, and at 27, 28, 69, and 70 DIM. The same 3 individuals recorded BCS during the entire experiment. In addition, calf birth weights and calving difficulty scores (1 = no problem; 2 = slight problem; 3 = needed assistance; 4 = considerable force; 5 = cesarean) were recorded by the attending herdsman. Births not attended were scored as 1. Cows were milked at 0600, 1400, and 2100 h, and milk yield was recorded daily. Samples for composition analysis were collected on d 7, 14, and 21 of lactation from all 3 milkings each day and were preserved using a tablet containing bronopol and natamycin (Broad Spectrum Microtabs II, D& F Control Systems, Inc., Dublin, CA). Incidences of milk fever, metritis, displaced abomasum, mastitis, and foot ailments were also recorded during the experiment.

Target day and actual day of blood sampling relative to calving were –21 and –20.8 (SD = 2.2), –14 and –14.6 (SD = 1.8), –7 and –8.0 (SD = 1.9), –2 and –2.5 (SD = 0.9), 2 and 2.2 (SD = 1.0), 7 and 7.0 (SD = 1.0), 14 and 14.1 (SD = 1.0), 21 and 21.3 (SD = 1.0), and 28 and 28.2 (SD =1.1), respectively. Approximately 4 h after feeding, blood was sampled from a coccygeal vessel into 2 evacuated tubes (Becton Dickinson and Co., Franklin Lakes, NJ) containing K-EDTA and sodium fluoride. Blood samples were immediately placed on ice and transported to the laboratory where they were centrifuged ($500 \times g$); plasma was harvested and stored at –20°C until analysis. Ruminal fluid was collected at 21 (SD = 5.4) and 7 (SD = 4.7) d before expected calving date and 7 (SD = 1.2) d postpartum 4 h after feeding by applying vacuum pressure to an esophageal tube fitted with a suction strainer. To minimize saliva contamination, approximately 250 mL of rumen fluid was discarded before sample collection. A 10-mL sample was mixed with 2 mL of 25% (wt/vol) metaphosphoric acid and frozen at –20°C until analyzed for concentrations of VFA and NH_3-N. Liver tissue was collected by trocar and aspiration between the 11th and 12th rib (Smith et al., 1997) approximately 4 h after feeding on d 7 and 14 [actual d were 7.2 (SD = 1.2) and 14.1 (SD = 1.0)] of lactation. Samples were blotted to remove any residual blood, split into 2 equal aliquots, placed into cryovials, immediately submerged in liquid nitrogen, and transported to the laboratory where they were frozen at –80°C until analysis.

Table 1. Ingredient composition of control (CON) and lactose (LAC) diets

Ingredient, % of diet DM	–21 to 0 DIM		1 to 21 DIM		22 to 70 DIM[1]
	CON	LAC	CON	LAC	
Brome grass hay	14.3	14.3	—	—	—
Alfalfa hay	14.5	14.5	8.3	8.3	15.3
Alfalfa haylage	—	—	14.5	14.5	6.6
Corn silage, processed	40.2	40.2	27.9	27.9	24.9
Whole cottonseed	2.0	2.0	8.6	8.6	7.9
Wet distillers grains	—	—	3.6	3.6	—
Dried distillers grains	—	—	—	3.8	4.6
Corn, high moisture	—	—	—	—	17.9
Corn grain, ground	17.7	—	25.2	5.4	7.6
Lactose[2]	—	15.7	—	15.7	—
Energizer 4-19W[3]	—	—	—	—	4.2
Soybean meal, 44%	1.81	1.65	4.84	4.30	2.43
SoyChlor 16-7[4]	6.63	6.64	—	—	—
SoyPlus[4]	1.76	3.60	4.26	4.77	4.51
Limestone	0.36	0.22	1.09	1.06	0.52
Dicalcium phosphate	—	0.18	0.19	0.33	—
Sodium bicarbonate	—	—	0.59	0.59	0.57
Fish meal, menhaden	—	—	—	—	0.40
Pork meat and bone meal	—	—	—	—	1.21
Yeast culture[5]	—	—	—	—	0.21
Magnesium oxide	—	—	0.24	0.24	0.17
Salt	—	—	0.24	0.24	0.50
Vitamin A, D, and E premix	0.65[a]	0.65[a]	0.31[b]	0.31[b]	0.31[b]
Urea	0.02	0.29	0.02	0.24	0.10
4-Plex[6]	0.07	0.07	0.07	0.07	0.05
Vitamin E premix[7]	—	—	0.05	0.05	0.02

[1]All cows were fed the same diet from 22 to 70 DIM.

[2]First District Ag Service, Litchfield, MN.

[3]Quality Liquid Feeds, Inc., Dodgeville, WI. Liquid mixture of cane molasses, condensed whey, and tallow (assay DM basis: 12.9% CP, 61% fat).

[4]West Central Soy, Ralston, IA. SoyChlor 16-7 is a heat-processed soybean meal treated with HCl (assay DM basis: 23% CP, 53% RUP, 10.3% Cl). SoyPlus is a heat-processed soybean meal (assay DM basis: 50% CP, 60% RUP).

[5]Diamond V XP, Diamond V Mills, Inc., Cedar Rapids, IA. *Saccharomyces cerevisiae* yeast and the media on which it was grown.

[6]4-Plex, Zinpro Corp., Eden Prairie, MN. Zn and Mn methionine complex, Cu lysine complex, and Co glucoheptonate.

[7]Contains 44,092 IU of vitamin E per kg.

[a]Contains 454,000 IU of vitamin A, 90,900 IU vitamin D, and 3,636, IU of vitamin E per kg.

[b]Contains 909,000 IU of vitamin A, 182,000 IU vitamin D, and 2,424, IU of vitamin E per kg.

Laboratory Analyses

Samples of diets were collected once weekly following delivery of the TMR to the Calan feed box. These samples were immediately dried at 55°C in a forced-air oven and allowed to air-equilibrate before being ground to pass a 2-mm screen of a standard Wiley mill (model 3, Arthur H. Thomas Co., Philadelphia, PA). Samples were composited by diet and month and analyzed by Dairyland Laboratories (Arcadia, WI) for DM at 105°C for 24 h, CP (AOAC, 1997) using a LECO-428 combustion analyzer (LECO Corp., St. Joseph, MI), NDF (Van Soest et al., 1991), ether extract (AOAC, 1997), minerals (AOAC, 1997; method 985.01), and ADF (AOAC, 1997). Determination of ADF was according to AOAC (method 973.18 C; 1997) whereas NDF was according to Van Soest et al. (1991) with the addition of 4 mL of α-amylase and 20 g of sodium sulfite using the Ankom A200 (Ankom Technology Corp., Fairport, NY) filter bag technique. Starch was measured as dextrose after treating samples with glucoamylase using a YSI 2700 Select Biochemistry Analyzer (Yellow Springs, OH; Holm et al., 1986). Minerals were quantified according to AOAC methods (985.01; 1997) using an inductively coupled plasma spectrometer (Thermo Jarrell Ash, Franklin, MA). Samples were also analyzed for lactose in our laboratory according to AOAC (method 974.06; 1990) using an HPLC (Waters Corporation, Milford, MA) equipped with a refractive index detector and a 300 × 7.8 mm column (HPX-87H, BioRad Laboratories, Hercules, CA) using a flow rate of 0.6 mL/min of 0.01 *N* H_2SO_4.

Milk compositional analysis was conducted by Heart of America DHI Laboratory (Manhattan, KS) according to approved procedures of AOAC (1990). Samples were composited by day and analyzed for true protein, fat, lactose (near infrared spectroscopy; Bentley 2000 Infrared Milk Analyzer, Bentley Instruments, Chaska, MN), MUN (a modified Berthelot reaction; ChemSpec 150 Analyzer, Bentley Instruments), and SCC (flow cytometer laser (Somacount 500, Bentley Instruments). The SNF was calculated.

Plasma samples were thawed and concentrations of glucose were determined using glucose oxidase (kit #315, Sigma Diagnostics, St. Louis, MO) according to the procedures of Trinder (1969). Concentration of BHBA in plasma was determined (Pointe Scientific, Inc., Lincoln Park, MI) following the methods of Williamson et al. (1962) and plasma NEFA concentrations were determined using a colorimetric assay (NEFA-C Kit, Wako Chemicals, Richmond, VA), following modifications by Johnson and Peters (1993). Insulin was quantified by solid-phase radioimmunoassay (Coat-A-Count, Diagnostic Products Corp., Los Angeles, CA) with an intraassay coefficient of variation of 1.6%. Samples of liver tissue were thawed and analyzed for total lipid and glycogen as described by Mills et al. (1986) and Derling et al. (1987), respectively.

Table 2. Nutrient composition of control (CON) and lactose (LAC) diets[1]

	–21 to 0 DIM		1 to 21 DIM		22 to 70
Nutrient	CON	LAC	CON	LAC	DIM[2]
DM, % as fed	49.3	55.1	54.2	55.0	48.7
CP, %	14.2	14.4	18.1	18.1	17.8
NE_L,[3] Mcal/kg	1.63	1.68	1.70	1.68	1.68
ADF, %	24.0	24.5	21.1	20.1	19.2
NDF, %	37.3	36.3	32.6	31.9	31.7
NFC,[4] %	37.0	36.7	37.2	37.3	36.5
Ether extract, %	4.3	5.3	5.5	4.6	6.6
Starch, %	25.8	12.6	25.7	10.5	29.0
Lactose, %	—	15.8	—	16.0	0.7
Ash, %	7.2	7.3	6.6	8.1	7.4
Ca, %	0.99	1.01	1.20	1.27	1.12
P, %	0.38	0.39	0.42	0.43	0.48
Mg, %	0.50	0.53	0.50	0.49	0.46
K, %	1.25	1.23	1.39	1.29	1.36
Na, %	0.16	0.17	0.35	0.36	0.48
Cl, %	0.89	1.09	0.33	0.41	0.58
S, %	0.21	0.20	0.27	0.27	0.29
DCAD,[5] mEq/kg of DM	8	–43	247	203	212

[1]Values are based upon nutrient analyses of diets and actual mean DMI by treatment.

[2]All cows were fed the same diet from 22 to 70 DIM.

[3]Calculated using NRC (2001).

[4]NFC = 100 – (% NDF + % CP + % ether extract + % Ash).

[5]DCAD as [(Na + K) – (Cl + S)] in milliequivalents per kg of DM.

Statistical Analyses

Effect of treatment on incidence of health disorders was not analyzed for statistical differences because of insufficient replication; however, all cows experiencing disorders were included in the data set. Two cows fed CON and 3 cows fed LAC gave birth to twins, all of which were natural deliveries. Of the cows fed CON that twinned, one was diagnosed with metritis at 10 DIM and the other was treated for a retained placenta. None of the cows fed LAC that twinned received any treatment for either metritis or retained placenta. One cow from each treatment was treated for mastitis and recovered quickly.

Milk yield and DMI data were reduced to weekly means for statistical analysis. Milk production data collected on the day of calving was not included in the statistical analysis. Data were analyzed as repeated measures using PROC MIXED (Littell et al., 1996) of SAS software, version 8.01 (2001). For each variable, cow was subjected to 4 covariance structures: autoregressive order 1, toeplitz, variance component, and compound symmetry. In general, the structure yielding the Akaike's information criterion closest to zero was variance component and autoregressive order 1. For variables measured over time, the model included treatment, time (week or day depending on the variable), and 2-way interactions as fixed effects. The random effect was diet nested within cow. The method of Kenward-Rogers was used for calculation of denominator degrees of freedom for *F*-tests. Covariates of initial BW and BCS, days on treatment, and previous 305-d mature equivalent milk yield were included for all data sets. Covariates and interactions were dropped from the model one at a time, starting with the least significant, and continuing until all remaining variables were significant. Prepartum and postpartum data were analyzed separately.

Statistical significance was declared at $P < 0.05$, with trends noted at $P > 0.05$ to $P < 0.15$. Least squares means and SEM are reported for all data. Significance of interactions is reported when significant. When significant effects between dietary treatments existed, mean separation was conducted by the PDIFF option in SAS.

RESULTS

Ingredient and nutrient composition of diets are shown in Tables 1 and 2, respectively. Diets were formulated using Cornell Penn Miner (CPM) Dairy (version 3.0.5; Cornell University, Ithaca, NY; University of Pennsylvania, Kennett Square, PA; William H. Miner Agricultural Research Institute, Chazy, NY) and recommendations from NRC (2001). A BW of 680 kg and

Table 3. Previous 305-d mature equivalent (ME) milk, days on treatment, BW, BCS, calf birth weights, calving difficulty, and prepartum DMI of cows fed control (CON) and lactose (LAC) diets

Item	Diet		SEM	P^1
	CON	LAC		
n	12	12	—	—
Previous 305-d ME, kg	12,254	11,010	467	0.07
Days on treatment	41.9	39.0	1.5	0.19
Lactation number[2]	1.8	1.7	0.26	0.66
BW,[3] kg				
d –21	777	775	11.7	0.92
d 0	704	702	11.7	0.92
d 28	657	643	12.3	0.43
d 70	678	669	12.7	0.61
BCS[4]				
d –21	3.34	3.47	0.06	0.17
d 0	3.33	3.20	0.06	0.12
d 28	3.02	2.89	0.07	0.16
d 70	3.09	3.05	0.07	0.71
Calf BW, kg	41.8	40.7	2.55	0.75
Calving difficulty[5]	1.4	1.8	0.28	0.36
Prepartum DMI, kg/d	12.2	13.3	0.59	0.22

[1]Significance of *F*-test.

[2]Lactation number before calving.

[3]Data represents means of d –22 and –21, 0 and 1, 27 and 28, and 69 and 70, respectively.

[4]Wildman et al., 1982.

[5]Five-point scale: 1 = no assistance, 2 = slight problem, 3 = needed assistance, 4 = considerable force, and 5 = cesarean.

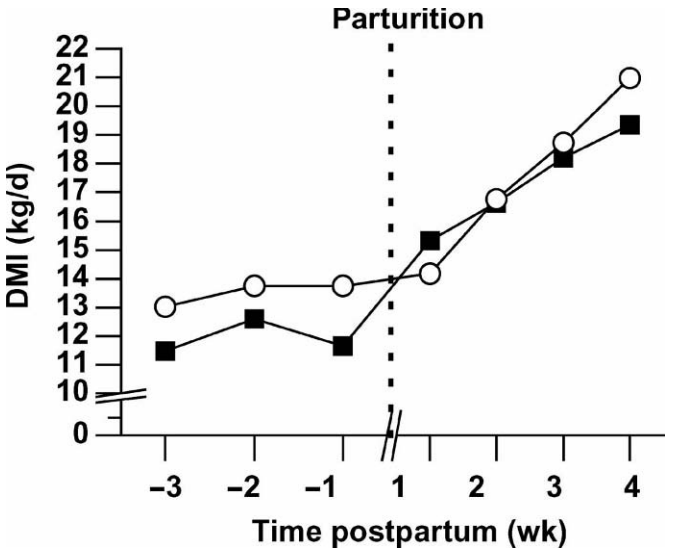

Figure 1. Dry matter intake (pooled SEM = 1.16) of cows fed control (■) and lactose (○) diets.

a targeted DMI of 12.2, 19.0, and 25.6 kg/d for pre-, postpartum, and lactation (29 to 70 DIM) diets, respectively, were used during formulations. The ratio of forage to concentrate was 70:30, 50:50, and 46:54 for pre-, postpartum, and lactation diets, respectively. The lactose content of LAC was 15.8% (SD = 0.79) and 16.0% (SD = 0.65) for the pre- and postpartum diets, respectively, and was slightly greater than the 15.7% formulated.

Initial cow characteristics are presented in Table 3. Average previous 305-d mature equivalent milk yield was 11,632 ± 467 kg. Previous 305-d mature equivalent yield tended ($P < 0.07$) to be lower for cows fed LAC. Therefore, previous 305-d milk yield was included as a covariate during statistical analysis of the data when significant. Days on treatment averaged 40.5 ± 1.5 d and number of lactations before initiation of treatments was 1.8 ± 0.26 and was similar among treatments. Initial BW and BCS were similar between treatments; however, BCS at parturition tended ($P = 0.12$) to be lower for cows fed LAC. Average prepartum DMI averaged 12.8 ± 0.59 kg/d and was not affected by diet. Dry matter intake decreased from wk 2 to wk 1 prepartum for cows fed CON, whereas intakes of cows fed LAC remained unchanged (Figure 1).

Postpartum performance data are presented in Table 4. Average DMI during the first 21 DIM was 17.7 ± 0.89 kg/d and did not differ between treatments. Yields of milk (Figure 2) and ECM (Orth, 1992) and production efficiencies (ECM/DMI) were not affected by dietary treatments during the first 21 DIM. Milk composition was not affected by dietary treatments. Body weight and BCS collected at 28 and 70 DIM also did not differ between treatments.

Effect of treatment on plasma metabolites are presented in Table 5 and plotted over time in Figures 3 and 4. When assessed as an overall mean of all samplings,

Table 4. Postpartum DMI, milk yield and composition, and milk SCC of cows fed control (CON) and lactose (LAC) diets

Item	Diet		SEM	P^1
	CON	LAC		
DMI, kg/d	18.2	17.2	0.89	0.43
Milk d 1 to 21, kg/d	40.7	38.6	2.46	0.54
ECM[2] d 1 to 21, kg/d	44.5	41.3	2.2	0.32
ECM/DMI d 1 to 21	2.97	2.60	0.28	0.37
Milk composition, d 1 to 21				
Fat, %	4.82	4.71	0.29	0.79
Fat kg/d	1.81	1.73	0.12	0.68
True protein, %	3.07	2.93	0.07	0.21
True protein, kg/d	1.17	1.07	0.07	0.36
SNF, %	8.42	8.42	0.13	0.99
SNF, kg/d	3.29	3.09	0.23	0.55
Lactose, %	4.63	4.72	0.11	0.55
Lactose, kg/d	1.86	1.69	0.12	0.33
SCC $\times 10^3$/mL	618	68	245	0.11
Urea N, mg/dL	11.41	11.33	0.72	0.94
Milk d 1 to 70, kg/d	44.8	46.6	2.13	0.57

[1]Significance of *F*-test.

[2]ECM = [(0.327 × kg milk) + (12.95 × kg fat) + (7.2 × kg protein)]; Orth, 1992.

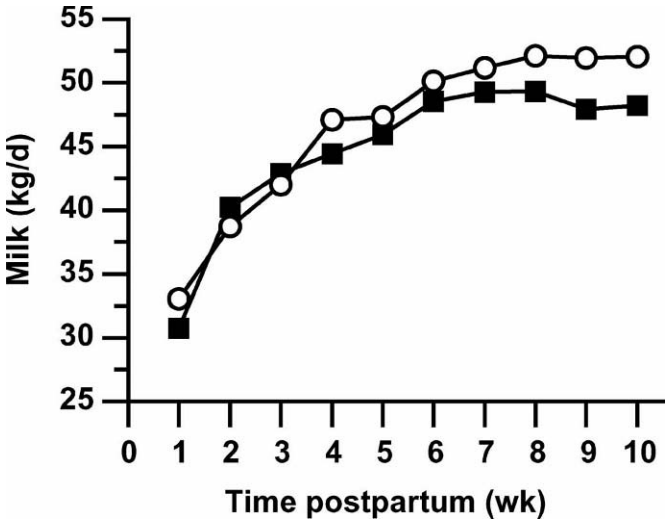

Figure 2. Milk yield (pooled SEM = 2.58) of cows fed control (■) and lactose (○) diets.

concentrations of plasma glucose and insulin were not different between treatments; however, differences were observed on specific days relative to calving. Concentrations of glucose in plasma were greater ($P < 0.05$) at 7 d prepartum (74.1 vs. 63.5 mg/dL) and tended ($P < 0.11$) to be greater at 14 d postpartum (66.7 vs. 59.6 mg/dL) in LAC compared with CON. Plasma insulin concentrations were greater at 21 (834 vs. 508 pg/mL) and 14 d (572 vs. 371 pg/mL) prepartum for cows fed LAC compared with those fed CON; however, postpartum concentrations tended ($P < 0.06$) to be greater (211 vs. 119 pg/mL) in cows fed CON at 21 DIM. Concentrations of NEFA in plasma were, on average, not different between treatments; although d 7 NEFA tended to be greater for LAC there were no significant diet × day interactions ($P > 0.72$). Concentrations of BHBA in plasma of cows fed LAC, however, were greater ($P < 0.01$) than for those fed CON both pre- and postpartum. These differences were most pronounced at 14 DIM when concentrations of BHBA in plasma were nearly 2-fold higher ($P < 0.01$) in LAC relative to CON (17.1 vs. 8.8 mg/dL). Dietary treatments did not alter liver glycogen content, which averaged 3.0% (wet weight; Table 5), but feeding LAC resulted in a 58% decrease

Table 5. Concentrations of glucose, insulin, NEFA, and BHBA in plasma and composition of livers of cows fed control (CON) and lactose (LAC) diets

Item	Diet		SEM	P^1
	CON	LAC		
Glucose, mg/dL				
All data	66.0	70.0	2.28	0.23
Prepartum	68.9	74.1	3.64	0.32
Postpartum	63.4	66.4	2.02	0.31
Insulin, pg/mL				
All data	258	319	37	0.26
Prepartum	367	526	84	0.19
Postpartum	170	145	12	0.18
NEFA, μEq/L				
All data	367	419	37	0.33
Prepartum	167	175	24	0.83
Postpartum	539	612	54	0.35
BHBA, mg/dL				
All data	7.65	10.06	1.02	0.07
Prepartum	4.25	6.14	0.31	<0.01
Postpartum	8.34	14.61	1.67	0.01
Liver glycogen, % wet weight	3.12	2.98	0.41	0.81
Liver lipid, % wet weight	14.7	8.6	1.49	0.01

[1]Significance of *F*-test.

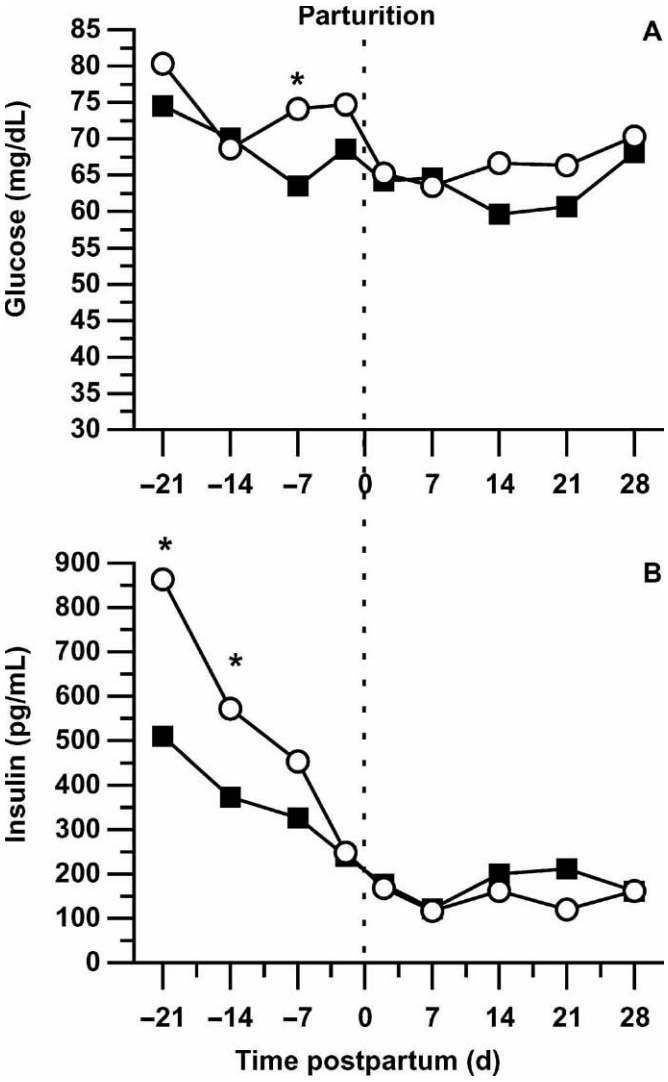

Figure 3. Plasma glucose (A) and insulin (B) concentrations (pooled SEM = 3.96 and 78.8, respectively) of cows fed control (■) and lactose (○) diets. Differences at individual times are indicated by * ($P < 0.05$).

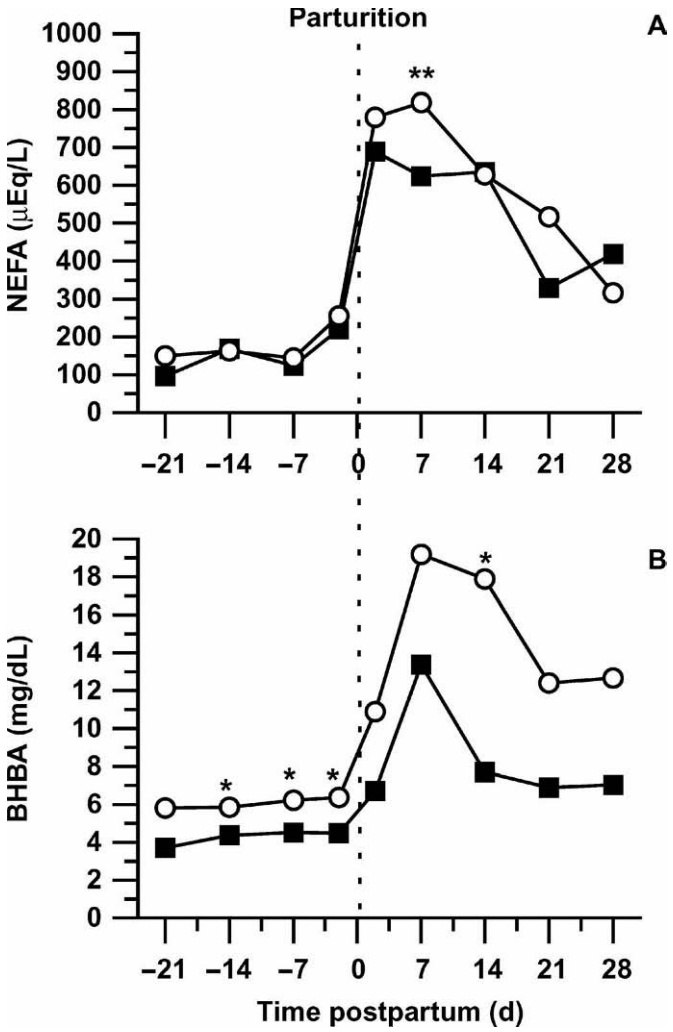

Figure 4. Plasma NEFA (A) and BHBA (B) concentrations (pooled SEM = 94 and 2.47, respectively) of cows fed control (■) and lactose (○). Differences at individual times are indicated by * ($P < 0.05$) and ** ($P < 0.10$).

($P < 0.01$) in liver lipid content relative to cows fed CON (8.6 vs. 14.7% wet weight).

Effects of diets on concentrations of NH_3-N and relative ruminal proportions of VFA are presented in Table 6. The ruminal NH_3-N concentrations were greater ($P < 0.04$) prepartum for CON compared with LAC but did not differ postpartum. Cows fed CON tended to have greater proportions of rumen propionate prepartum and significantly greater propionate postpartum ($P < 0.04$.). Proportions of butyrate in rumen VFA increased ($P < 0.01$) an average of 2.4 percentage units pre- and postpartum for cows fed LAC. The proportion of rumen VFA as branched-chain fatty acids decreased by 0.85 percentage units in cows fed LAC relative to those fed CON. Total concentrations of VFA, the proportion of acetate, and the ratios of acetate to propionate were not affected by treatments.

Table 6. Ruminal NH_3-N and VFA proportions of cows fed control (CON) and lactose (LAC) diets

	Diet			
Item	CON	LAC	SEM	P^1
Prepartum				
NH_3-N, mg/dL	3.25	2.37	0.30	0.04
Total VFA, m*M*	46.4	50.5	4.29	0.50
VFA, molar proportions				
Acetate	68.3	67.8	0.77	0.66
Propionate	19.6	18.8	0.38	0.15
Isobutyrate	0.80	0.44	0.13	0.06
Butyrate	9.2	11.3	0.45	<0.01
Isovalerate	1.14	0.61	0.08	<0.01
Valerate	1.05	1.09	0.10	0.78
Branched-chain fatty acids	1.94	1.05	0.17	<0.01
Acetate:propionate	3.52	3.64	0.10	0.42
Postpartum				
NH_3-N, mg/dL	2.69	2.29	0.48	0.56
Total VFA, m*M*	40.8	34.3	4.58	0.33
VFA, molar proportions				
Acetate	63.7	64.7	2.8	0.62
Propionate	22.5	19.5	0.98	0.04
Isobutyrate	1.04	0.78	0.19	0.33
Butyrate	10.3	13.0	0.67	0.01
Isovalerate	1.24	0.68	0.22	0.08
Valerate	1.16	1.39	0.20	0.43
Branched-chain fatty acids	2.28	1.43	0.40	0.16
Acetate:propionate	2.98	3.38	0.25	0.28

[1]Significance of *F*-test.

DISCUSSION

Successful transition into lactation is highly influenced by maintaining feed intake during the periparturient period. Prepartum DMI is inversely related to concentrations of NEFA and BHBA in plasma as well as liver triglyceride content (Bertics et al., 1992). In this experiment, feeding lactose did not affect DM consumed; however, DMI patterns across time were distinctive. Cows fed LAC did not exhibit a prepartum depression in DMI (Figure 1) and consumed 13.5 kg of DM from 3 wk before calving through 7 DIM, whereas cows fed CON exhibited feed intake depression (nadir = 11.6 kg/d) 1 wk before calving. DeFrain et al. (2004) reported trends for greater DMI in cows fed diets containing up to 14% of DM as lactose, whereas others have found no effect of lactose on DMI (Doreau et al., 1987; Maiga et al., 1995).

Analysis of the magnitude of intake depression indicated no differences between treatments; however, the intake patterns observed invite postulation. The absence of a depression in DMI (Figure 1) suggests that lactose may have improved rumen function and therefore nutrient supply and use. Allen and Xu (1998) demonstrated increased length, width, and surface area of

ruminal papillae in nonpregnant, nonlactating dairy cows fed diets containing 43% lactose (DM basis) compared with 43% corn. Concentrations of rumen VFA were not reported by Allen and Xu (1998); however, there are reports of increased proportions of butyrate in rumens of cows fed diets containing lactose (Schingoethe, 1976; DeFrain et al., 2004). Butyrate has been shown to stimulate papillae development and growth resulting in increased surface area and absorptive capacity (Dirksen et al., 1985).

The absence of treatment effects on milk composition by feeding lactose or whey has been observed (Schingoethe, 1976). Other researchers found lactose and whey products to maintain or slightly increase milk fat when substituted for ground corn (Schingoethe et al., 1976; Bowman and Huber, 1967) presumably because the ruminal butyrate and plasma BHBA contribute to fatty acid synthesis in the mammary gland (Palmquist et al., 1969). The ability of lactose and whey products to affect milk fat may be dependent upon diet presentation and stage of lactation. Cows in our study were fed a TMR and were less than 21 DIM, whereas others have used component-fed diets (Bowman and Huber, 1967; Schingoethe et al., 1976) or later lactation cows (>100 DIM; Schingoethe et al., 1976; DeFrain et al., 2004).

Feeding lactose did not affect protein in milk, which agreed with results from Pinchasov et al. (1982) and DeFrain et al. (2004) but not Schingoethe et al. (1976) who found substituting dried whole whey (5% diet DM) for shelled corn during late lactation (180 DIM) increased milk protein percentage over control-fed cows (3.95 and 3.80%, respectively). Likewise, feeding lactose did not affect MUN. This is contrary to known effects of lactose on N efficiency (Poncet and Rayssiguier, 1980; King and Schingoethe, 1983), particularly because rumen NH_3-N tended to be decreased in cows fed LAC prepartum but not postpartum. Feeding lactose has been observed to decrease concentrations of urea N in milk and NH_3-N in rumen fluid when substituted for cornstarch (DeFrain et al., 2004). Relative to unsupplemented or corn and soybean meal-based controls, feeding lactose has decreased concentrations of rumen NH_3-N in both high-forage (Poncet and Rayssiguier, 1980) and high-concentrate (King and Schingoethe, 1983) diets. Decreased ruminal NH_3-N and MUN suggest there may have been an opportunity for greater ruminally degradable protein supply and therefore greater microbial protein synthesis from lactose as previously reported by Susmel et al. (1995).

The objectives of the experiment were met as the fermentation of lactose increased proportions of butyrate in rumens and BHBA in blood of transition dairy cows. Feeding lactose has consistently increased rumen butyrate (Schingoethe, 1976; DeFrain et al., 2004) and plasma BHBA (Doreau et al., 1987; DeFrain et al., 2004). According to Weigand et al. (1975), the enzyme system involved in rumen epithelial ketogenesis may become saturated; however, changes in concentrations of butyrate in plasma were undetectable in cows fed lactose at 14% of the diet DM (DeFrain et al., 2004). Our hypothesis, supported by Black et al. (1966) and Anand and Black (1970), was that increases in plasma BHBA would increase hepatic gluconeogenesis via allosteric activation of pyruvate carboxylase by the presence of acetyl CoA units created from BHBA metabolism. In our experiment, concentrations of BHBA in plasma were greater for cows fed LAC, especially during 7 to 21 DIM, a time when pyruvate carboxylase mRNA expression is greatest (Greenfield et al., 2000). Increased concentrations of BHBA in plasma could have resulted from increased alimentary ketogenesis, decreased uptake of ketone bodies by tissues, or increased partial oxidation of fatty acids in livers. The latter is supported by the observation that concentrations of NEFA in plasma were similar between treatments yet total lipid content of livers was decreased for cows fed LAC. Concentrations of plasma BHBA peaked for all cows at 7 DIM and remained elevated through 14 DIM for cows fed LAC compared with cows fed CON. During this same period (7 to 14 DIM), concentrations of glucose in plasma decreased in cows fed CON and increased (remaining stable through 28 DIM) for cows fed LAC (Figure 3), supporting the hypothesis that BHBA in plasma may have up-regulated gluconeogenic enzymes. Further evidence of this effect is that the elevated concentrations of glucose in blood was not at the expense of hepatic glycogenolysis as glycogen content of livers was similar between treatments. DeFrain et al. (2004) found that glucose in plasma was decreased from 68.0 to 65.5 mg/dL when lactose was fed at 14% of diet DM. Differences between studies may be a result of differences in stage of lactation, which is known to affect the expression of hepatic gluconeogenic enzymes (Greenfield et al., 2000).

Although glycogen content was unaffected by treatments, lipid content in livers of cows fed LAC was decreased by nearly 40% relative to those fed CON. Indicators of lipolysis (BW and BCS losses and concentrations of NEFA in plasma), however, were unaffected by dietary treatments. Therefore, it is possible that the increase in plasma BHBA observed in cows fed LAC in the current study mediated the effects of treatments on liver lipid content and metabolism. One possibility is that elevated BHBA in blood of cows fed LAC increased the efficiency of assembly or secretion of liver triglycerides as very low density lipoproteins, although the authors are unaware of any documentation of ketone bodies as regulators of lipoprotein synthesis and

export. A second explanation is that, because of the similarities in postpartum plasma NEFA profiles (Figure 4), the increased BHBA in blood of cows fed LAC may have been partially attributable to increased ability of the liver to oxidize fatty acids to ketone bodies. A review on ketone body use in ruminants by Heitmann et al. (1987) reported BHBA infusions in sheep stimulated insulin secretion and production by the pancreas, decreased NEFA, decreased hepatic uptake of NEFA and ketogenesis. Therefore, effects of feeding LAC on liver lipid content may have also been related to decreased hepatic uptake of NEFA. This scenario is supported by the transient increase in blood NEFA at 7 DIM.

CONCLUSIONS

Feeding lactose from 21 d pre- to 21 d postcalving increased the proportions of butyrate in rumens and BHBA in blood of dairy cows. This increase in circulating ketones was offset by marginal increases in concentrations of blood glucose. Evidence of improved absorptive capacity of the rumen and nutrient use is provided by DMI remaining constant during the 3 wk before calving and improvements in rumen NH_3-N use. Of great interest is an observed decrease in liver lipid concentrations. These data indicate that substituting lactose for corn is a viable option for producers with access to lactose-containing products such as whey.

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