

Impact of *Saccharomyces cerevisiae* Fermentation Product on Feed Intake Parameters, Lactation Performance, and Metabolism of Transition Dairy Cattle

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Summary

A *Saccharomyces cerevisiae* fermentation product (NutriTek; Diamond V, Cedar Rapids, IA) was fed from 29 ± 5 days before calving and through 42 days in milk (DIM) to evaluate the effects on feed intake parameters, milk production, and metabolism. Treatments were control ($n = 30$) or 18 g/d NutriTek (NT, $n = 34$) provided as total mixed rations. Cows were individually fed 3 \times /day prepartum and 2 \times /day postpartum. Cows were milked 2 \times /day with samples taken 2 \times /week for composition analysis. Body weight (BW) was measured at enrollment (day -29 ± 5), day 0, and day 42 relative to calving, and body condition was scored weekly. Blood samples were collected during weeks -4, -2, 1, 2, and 5 relative to calving for biomarkers of metabolism and inflammation. To evaluate adaptive immunity, cows were challenged with a subcutaneous injection of ovalbumin (egg protein) and immune response was determined by serum concentrations of anti-ovalbumin immunoglobulin G (IgG) on days 7, 21, 28, and 35 of lactation. Overall dry matter intake, BW, body condition score, and milk yield were not different between treatments. NutriTek did alter feeding behavior by increasing the number of meals consumed with less time between those meals. Milk fat concentration increased with NT during weeks 4 and 5 of lactation, which contributed to an increase in fat yield during those weeks. There were tendencies for greater milk lactose yield in control cows and greater milk urea nitrogen concentration in NT, but no treatment differences for milk protein concentration or somatic cell count. Assuming equal digestibility, energy balance was more negative for NT during weeks 4 and 5, mirroring the increase in milk fat during that time. Energy density of diets calculated from observed ECM yield and BW change did not differ by treatment. Plasma concentrations of free fatty acids, β -hydroxybutyrate (BHB), glucose, insulin, and the inflammation marker haptoglobin did not differ between treatments. NutriTek increased the incidence of subclinical ketosis (12 vs. 38%, diagnosed by urine ketones). There was no overall treatment effect for immune response to vaccination; however, a treatment \times parity interaction indicated greater antibody concentration in primiparous cows supplemented with NT. A partial budget analysis accounting for milk income, feed cost, and expense associ-

¹Diamond V, Cedar Rapids, IA.

ated with ketosis treatment indicated an additional \$0.35 daily profit per cow for NT vs. control. In conclusion, NT supplementation during the transition period altered feeding behavior and milk fat concentration and ultimately appeared profitable in this scenario, despite the increased incidence of subclinical ketosis and a lack of response in early lactation milk yield.

Introduction

Saccharomyces cerevisiae fermentation products have been reported to influence the rumen environment, increasing fiber digestion, lactic acid utilization, and rumen pH. These attributes may be particularly advantageous during periods of stress, and may explain why dietary *Saccharomyces cerevisiae* fermentation product increases dry matter intake in early lactation by an average of 1.37 lb/day and energy-corrected milk by 3.64 lb/day. Beyond these proposed effects of *Saccharomyces cerevisiae* fermentation products on ruminal health, they may also affect transition cows through altered feeding behavior and immune function.

The objective of this study was to determine the effects of a new *Saccharomyces cerevisiae* fermentation product (NutriTek, Diamond V, Cedar Rapids, IA) on feed intake, feeding behavior, milk production and composition, energy balance, metabolism, and adaptive immunity during the transition period in dairy cows.

Experimental Procedures

Sixty-four parturient Holstein cows (50 multiparous, 14 primiparous) were used in a randomized block design. Cows were blocked by parity, expected calving date, and previous 305ME yield, then randomly assigned to treatment within block. Treatments were either control ($n = 30$) or 18 grams NT per day ($n = 34$) that was incorporated into a total mixed ration (TMR). Treatments were fed from -29 ± 5 to 42 days relative to calving. Feed ingredient samples were collected once weekly, composited by 4 months, and analyzed by wet chemistry methods for dry matter, neutral detergent fiber, starch, crude protein, ether extract, and ash content (Dairy One, Ithaca, NY). Chemical analyses of individual feed ingredients were used for determination of TMR nutrient composition (Table 1).

Parturient cows were fed treatment diets using an electronically gated feeding system (Roughage Intake System; Insentec B.V., Marknesse, the Netherlands). All cows on a given treatment diet were allowed access to 4 feed bins assigned to that treatment, and no more than 6 animals shared those 4 bins at any given time. To account for the capacity of the feed bins and potential bunk favoritism, parturient cows were fed 3×/day. Upon calving, cows were moved to a tie-stall facility where they were fed individually twice daily. Both feeding systems electronically recorded individual feed consumption and meal patterns. As-fed feed intake was recorded on a daily basis and adjusted by TMR dry matter for determination of meal and daily dry matter intake (DMI). For feeding behavior analysis, the inter-meal interval (IMI) was defined by a gap of at least 12 minutes between distinct meals and the minimum meal weight was 0.9 lb.

Cows were milked 2×/day with milk weights recorded for each milking. Milk samples were collected 2×/week and analyzed for concentrations of fat, true protein, lactose

(B-2000 Infrared Analyzer; Bentley Instruments, Chaska, MN), milk urea nitrogen (MUN; MUN spectrophotometer, Bentley Instruments), and somatic cells (SCC 500, Bentley Instruments) by MQT Laboratories (Kansas City, MO). Energy-corrected milk was calculated as $(0.327 \times \text{milk yield}) + (12.95 \times \text{fat yield}) + (7.65 \times \text{protein yield})$, and fat-corrected milk (FCM) was calculated as $(0.432 \times \text{milk yield}) + (16.216 \times \text{fat yield})$.

Body condition score (BCS) was recorded weekly by 3 trained investigators. Body weight was measured at enrollment (-29 ± 5 days relative to calving), after calving, and at 42 DIM. Prepartum energy balance was calculated as net energy (NE) intake – (NE maintenance + NE pregnancy). A total of 5 jugular blood samples were taken from each cow on the following weeks relative to calving: -4, -2, 1, 2, and 5. Bloods samples were analyzed for concentrations of free fatty acids, β -hydroxybutyrate, glucose, insulin, and haptoglobin. To evaluate adaptive immunity, cows were challenged with a subcutaneous injection of ovalbumin and Quil-A adjuvant on days 7 and 21 postpartum. Immune response was measured by anti-ovalbumin IgG concentration in serum samples collected on days 7, 21, 28, and 35 postpartum.

Cow health was evaluated daily by visual inspection, rectal temperature, and urine acetoacetic acid concentration (KetoCare, TRUEplus). Cows were monitored for disorders including ketosis (urine acetoacetic acid concentration > 40 mg/dL), milk fever, displaced abomasum, retained placenta, metritis, and mastitis. All diagnosed disease and health issues were recorded.

Results and Discussion

Dry Matter Intake and Feeding Behavior

Dry matter intake exhibited the typical decrease as calving approached and increased with the progression of lactation (Figure 1A); however, DMI was not altered by NT supplementation ($P > 0.69$). Similar to DMI, all feeding behavior parameters except IMI ($P = 0.28$) were influenced by day relative to calving ($P < 0.01$; Table 2). NutriTek tended to increase prepartum meal count ($P = 0.06$; Figure 1B) and decreased the time between meals ($P = 0.03$; Figure 1C), specifically during the 10 days preceding calving. A treatment \times week interaction for meal weight ($P = 0.03$) indicated control cows consumed larger meals during days -7 to -4 relative to calving. These data suggest that NT cows consumed lighter meals more often with less time between those meals leading up to calving. Interestingly, the treatment \times parity interaction for meal count and IMI ($P \leq 0.03$) suggested this altered feeding behavior with NT mainly applied to primiparous cows. Meal count was greater (9.7 vs. 8.5 ± 0.4 meals/day) and IMI lesser (0.65 vs. 0.82 ± 0.04 hours) for primiparous NT cows, but both were similar for multiparous NT and control cows (8.8 vs. 8.9 ± 0.2 meals/day and 0.82 vs. 0.81 ± 0.02 hours). Postpartum, NT cows continued to consume more meals ($P = 0.03$) with a tendency for less time between meals ($P = 0.07$). Considering the lesser proportion of primiparous cows in this study, the prepartum treatment \times parity interaction for meal count could partially explain why we observed differences in feeding behavior that did not translate into greater DMI. There was no treatment \times parity interaction ($P > 0.20$) for postpartum meal count, therefore this potential explanation would not extend after calving.

These modulations to feeding behavior have been documented in previous transition cow studies supplementing *Saccharomyces cerevisiae* fermentation product. The more consistent meal patterns may contribute to improved, more stable rumen function in NT-fed cows.

Body Weight, Body Condition, Milk Production, and Energy Balance

Cows experienced the typical decrease in body condition and body weight during the transition to lactation ($P < 0.001$); however, there was no effect of treatment or treatment \times time interaction for either ($P > 0.50$). On average, cows lost 0.7 BCS units (3.6 to 2.9) and 196 lb of BW (1,519 to 1,323 lb) during the experiment.

As shown in Table 3 and Figure 2A, most milk production parameters (milk yield, energy-corrected milk, and fat-corrected milk) were not affected by treatment ($P \geq 0.32$). Milk fat concentration increased ($P = 0.01$) and milk fat yield tended to be greater ($P = 0.10$; Figure 2B) for NT cows, with differences in weeks 4 and 5. We observed no differences for milk protein yield and content, lactose yield, and milk somatic cell linear score ($P > 0.15$). Milk lactose concentration tended to be greater for control ($P = 0.06$) and MUN tended to be greater for NT ($P = 0.06$). Greater milk fat content in transition cow studies could indicate greater release of body fat; however, lack of difference in BCS and timing of the milk fat response (week 4 and 5) make that unlikely. Risk of ruminal acidosis is increased during the postpartum period. It is possible the more consistent meal patterns contributed to improved rumen function, which could decrease the risk for shifts in biohydrogenation pathways and milk fat depression. *Saccharomyces cerevisiae* fermentation product has also been documented to increase fiber-digesting bacterial populations that largely produce acetate, one of the main lipogenic precursors for de novo fatty acid synthesis. However, these potential mechanisms are unlikely to be involved only during weeks 4 and 5, making it a less compelling explanation. The exact mechanisms involved in the observed increase in milk fat yield are unclear.

Energy balance (calculated assuming equal nutrient digestibility across diets) differed by week ($P < 0.001$) and treatment ($P = 0.03$). The lesser energy balance for NT during weeks 4 and 5 aligns with the time of increased milk fat concentration. Energy balance was also less in primiparous compared to multiparous cows (-7.02 vs. -3.47 ± 0.83 Mcal/day; $P < 0.01$). To account for possible differences in nutrient digestibility, we calculated energy density of the diet as energy required (milk energy + maintenance) minus energy supplied from mobilized body reserves, divided by dry matter intake. This calculation of observed feed energy (observed diet NE_L concentration) could provide some insight into changes in digestible or metabolizable energy supply from the diet; however, no difference between diets was detected ($P = 0.18$).

Metabolic Signaling and Adaptive Immune Response

Changes over time for plasma free fatty acids, BHB, insulin, and glucose reflected the typical metabolic and endocrine changes during the transition period ($P < 0.001$). The metabolic profile was not altered by NT supplementation ($P > 0.35$; Table 4). Plasma haptoglobin concentration, a marker of inflammation, tended to differ by week ($P = 0.08$), but not by treatment ($P = 0.18$).

Potential effects of NT on the adaptive immune system were evaluated by the response to a subcutaneous injection of ovalbumin. Antibody production increased after the challenge ($P < 0.001$) as expected. There was no overall treatment effect ($P = 0.25$), but the treatment \times parity interaction indicated greater anti-ovalbumin IgG concentration in primiparous cows supplemented with NT compared to control (0.36 vs. 0.28 ± 0.08 optical density; $P = 0.08$). It is possible that NT enhanced B lymphocyte activation, thus increasing antibody production in primiparous cows.

Disease Incidence

Incidences of common periparturient diseases occurring throughout the study period are outlined in Table 5. No metabolic diseases - except ketosis - differed by treatment ($P > 0.10$). Incidence of subclinical ketosis (SCK) diagnosed via urine ketones was greater in cows supplemented with NT compared to control cows (38% vs. 12%; $P = 0.02$; Figure 4A) and days of glucogenic treatment were also greater (1.7 vs. 0.4 ± 0.3 d; $P = 0.01$). To understand the observed increase in SCK incidence, despite little evidence of an overall treatment effect on plasma ketone concentrations or decreased energy balance during the window of time when ketosis was observed, a deeper investigation was carried out. The majority of ketosis diagnosis occurred between 10 and 20 days in milk ($n = 8$, NT = 6, control = 2). Because of this timing, we used week 2 plasma data to explore potential mechanisms underlying this effect. First, urine diagnosis of ketosis by urine ketones was effective, as plasma BHB concentrations were clearly greater in cows diagnosed with SCK versus those that were not ($P < 0.001$). Analysis of week 2 BHB concentrations demonstrated a parity \times treatment interaction ($P = 0.02$; Figure 4B): treatment did not impact BHB in primiparous cows, but NT increased BHB concentrations in multiparous cows. The observed increase in BHB in multiparous cows fed NT occurred without any other signs of poor health – we did not observe any clinical signs, and feed intake was not impaired in this group. We suspect that the increased BHB production is a metabolic response to NT rather than a true sign of disease.

Partial Budget

A simple partial budget analysis was conducted incorporating milk income, feed costs, and the expense associated with treating ketosis (Table 6). Milk income was generated using milk fat and protein prices of \$2.51 and \$1.78 per lb, respectively. Individual feed ingredient costs represented those of August 2016. Prepartum and postpartum DMI and diet composition were used to calculate diet prices on a daily cow basis, and the cost of NT was incorporated at \$0.13 per cow daily. Ketosis cases were treated with propylene glycol (\$4/treatment) for 3 days, so ketosis incidence and treatment costs were then used to determine the ketosis expense on a daily per-cow basis for the 42-day postpartum period. This analysis revealed an additional \$0.35 profit per cow daily with NT supplementation. This analysis included all numerical differences between treatments; however, since DMI and production parameters other than milk fat concentration did not statistically differ between treatments, cautious interpretation is warranted.

Conclusions

Supplementation with NT during the transition period altered prepartum and postpartum feeding behavior, with increased meals per day and decreased time between

those meals. Although no effects were detected for DMI, milk yield, milk protein, or SCC, milk fat percent was increased by approximately 13% in cows receiving NT, with differences beginning after the time period in which lipid mobilization is greatest during the transition period. Body weight, BCS, energy metabolites, and an inflammatory biomarker were unaffected. Incidence of SCK was increased with NT, but the mechanism through which this occurred has yet to be elucidated. Despite the increased incidence of SCK and no observed effect in milk yield, the increased milk fat contributed to an increased marginal economic return for NT over the first 42 days of lactation.

Table 1. Ingredient and nutritional composition of the prepartum and postpartum diets of control cows and cows supplemented with a *Saccharomyces cerevisiae* fermentation product (NutriTek; NT) from -29 ± 5 days relative to calving through 42 DIM

	Prepartum		Postpartum	
	Control	NT	Control	NT
Ingredient, % of dry matter				
Alfalfa hay ¹	---		9.75	
Alfalfa hay ²	---		9.70	
Grass hay	38.03		1.62	
Corn silage	19.83		22.32	
Wet corn gluten feed ³	18.44		23.76	
Cotton seed	---		4.05	
Ground corn	8.28	8.23	17.92	17.87
Micronutrient premixes	15.43	15.48	10.88	10.94
Nutrient, % of dry matter (unless otherwise specified)				
Dry matter, % as-fed	63.3		59.7	
Crude protein	12.9		17.0	
Acid detergent fiber	25.0		17.8	
Neutral detergent fiber	43.1		31.3	
Nonfiber carbohydrates	30.1		37.6	
Starch	15.3		22.6	
Crude fat	5.1		6.3	
NE _L , Mcal/kg	1.42		1.66	

¹Lower quality alfalfa with 22.1% crude protein.

²Higher quality alfalfa with 23.9% crude protein.

³Sweet Bran (Cargill Inc., Blair, NE).

NE_L = net energy for lactation.

Table 2. Dry matter intake (DMI), water intake, and feeding behavior parameters for control cows and cows supplemented with a *Saccharomyces cerevisiae* fermentation product (NutriTek; NT) from -29 ± 5 days relative to calving through 42 days in milk

	Control	NT	SEM ¹	<i>P</i> -values				
				Treatment	Time ²	Treatment × time	Parity	Treatment × parity
Prepartum measure								
DMI, lb/day	25.40	25.84	1.10	0.70	< 0.001	0.76	< 0.01	NS
Meal count/day	8.66	9.27	0.22	0.06	< 0.001	0.44	0.52	0.03
Meal weight, lb	2.89	2.84	0.13	0.75	< 0.001	0.03	< 0.01	NS
Meal length, minutes	28.28	29.49	0.94	0.28	< 0.001	0.03	0.91	NS
Inter-meal interval, hours	2.26	2.09	0.05	0.03	0.28	0.10	0.04	0.01
Postpartum measure								
Water intake, L/day	104.3	109.7	3.7	0.16	< 0.001	0.60	< 0.001	0.32
DMI, lb/day	45.38	45.62	1.15	0.84	< 0.001	0.75	< 0.001	NS
Meal count/day	11.35	12.60	0.45	0.03	< 0.001	0.70	0.52	NS
Meal weight, lb	4.39	4.45	0.24	0.83	< 0.001	0.34	0.45	NS
Meal length, minutes	23.77	25.77	1.30	0.22	< 0.001	0.62	0.05	NS
Inter-meal interval, hours	1.81	1.62	0.09	0.07	< 0.001	0.55	0.07	NS

¹Pooled standard error of the mean.²Time is by week for DMI and by day relative to calving for feeding behavior parameters.**Table 3. Lactation performance and energy balance for control cows and cows supplemented with a *Saccharomyces cerevisiae* fermentation product (NutriTek; NT) from 29 ± 5 days prepartum to 42 days in milk**

	Control	NT	SEM ¹	<i>P</i> -values		
				Treatment	Week	Treatment × week
Milk, lb/day	91.18	88.71	2.78	0.43	< 0.001	0.24
Milk fat, %	3.96	4.32	0.11	0.01	< 0.001	< 0.05
Milk fat, lb/day	3.62	3.90	0.18	0.10	< 0.001	0.09
Milk protein, %	3.03	3.12	0.04	0.16	< 0.001	< 0.01
Milk protein, lb/day	2.73	2.67	0.09	0.48	< 0.001	0.61
Milk lactose, %	4.93	4.87	0.02	0.06	< 0.001	0.70
Milk lactose, lb/day	4.50	4.34	0.13	0.29	< 0.001	0.41
Milk urea nitrogen, mg/dL	11.51	12.42	0.38	0.06	< 0.001	0.21
Milk somatic cell linear score ²	2.32	1.94	0.28	0.29	< 0.001	0.55
Energy-corrected milk, lb/day	96.78	99.69	3.66	0.41	< 0.001	0.09
Fat-corrected milk, lb/day	96.80	101.28	4.19	0.32	< 0.001	0.20
Energy balance, Mcal/day	-4.34	-6.15	0.74	0.03	< 0.001	0.20
Observed diet NE _L , Mcal/kg DM	1.83	1.90	0.04	0.18	< 0.001	0.19

¹Pooled standard error of the mean.²SCLS = $\log_2(\text{somatic cell count}/100) + 3$.

Table 4. Metabolic and inflammatory biomarkers in plasma of control cows and cows supplemented with a *Saccharomyces cerevisiae* fermentation product (NutriTek; NT) from 29 ± 5 days prepartum to 42 days in milk

	Control	NT	SEM ¹	P-values		
				Treatment	Week	Treatment × week
Glucose, mg/dL	64.0	62.7	2.06	0.60	< 0.001	0.14
Insulin, µg/L	0.12	0.12	0.01	0.46	< 0.001	1.00
NEFA, ² µEq/L	420	444	21	0.36	< 0.001	0.13
BHBA, ³ µM	556	572	22	0.57	< 0.001	0.12
Haptoglobin, ng/mL	514	575	42	0.18	0.08	0.51

¹Pooled standard error of the mean.

²Non-esterified fatty acids.

³Beta-hydroxybutyric acid.

Table 5. Disease incidence for control cows and cows supplemented with *Saccharomyces cerevisiae* fermentation product (NutriTek; NT) from 29 ± 5 days prepartum to 42 days in milk

	Control	NT
At-risk ¹	33	32
Fever	9	5
Displaced abomasum	0	2
Retained placenta	2	0
Ketosis	4	12 [*]
Mastitis	2	1
Other ²	4	1

¹Includes all cows that surpassed the exclusion criteria at calving. Cows excluded from analysis due to periparturient issues were included.

²Other includes 1 case of peritonitis resulting in death (control), 3 foot injuries (2 control, 1 NT), and 1 diarrhea/digestive upset at calving (control).

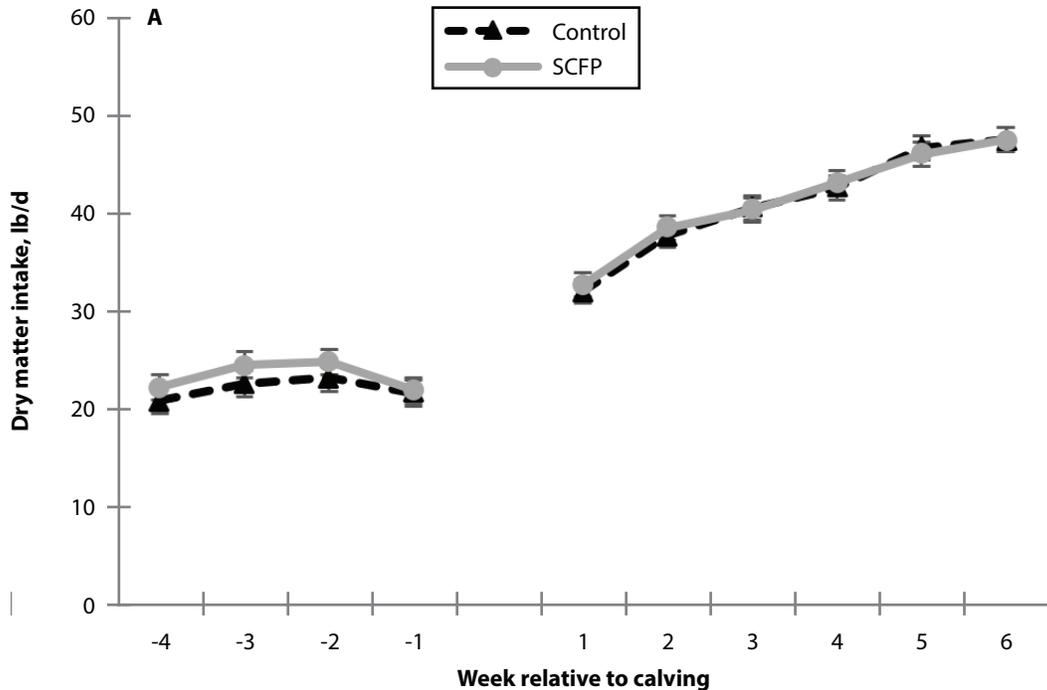
^{*}Fisher's exact test: $P = 0.02$. No other conditions were significantly affected by treatment.

Table 6. Partial budget analysis of supplementing cows with a *Saccharomyces cerevisiae* fermentation product (NutriTek: NT) from 28 days before calving through 42 days in milk

Revenue or expense category	Control	NT
Milk income		
Fat income per cow, \$2.51/lb	\$9.08	\$9.80
Protein income per cow, \$1.78/lb	\$4.87	\$4.75
	\$13.94	\$14.55
Feed cost¹		
Prepartum per cow/day	\$2.67	\$2.85
Postpartum per cow/day	\$5.45	\$5.61
	(\$5.70)	(5.87)
Expense due to ketosis (per cow daily over 42 days at risk)		
Ketosis incidence, %	12	38
Cost of treatment per case, ² \$	\$12.00	\$12.00
	(\$0.03)	(\$0.11)
Income over feed and treatment costs	\$8.22	\$8.57

¹Overall feed cost divides prepartum feeds costs over 305 days plus postpartum feed costs.

²Ketosis treatment cost represents 3 days of propylene glycol at \$4/day, including labor.



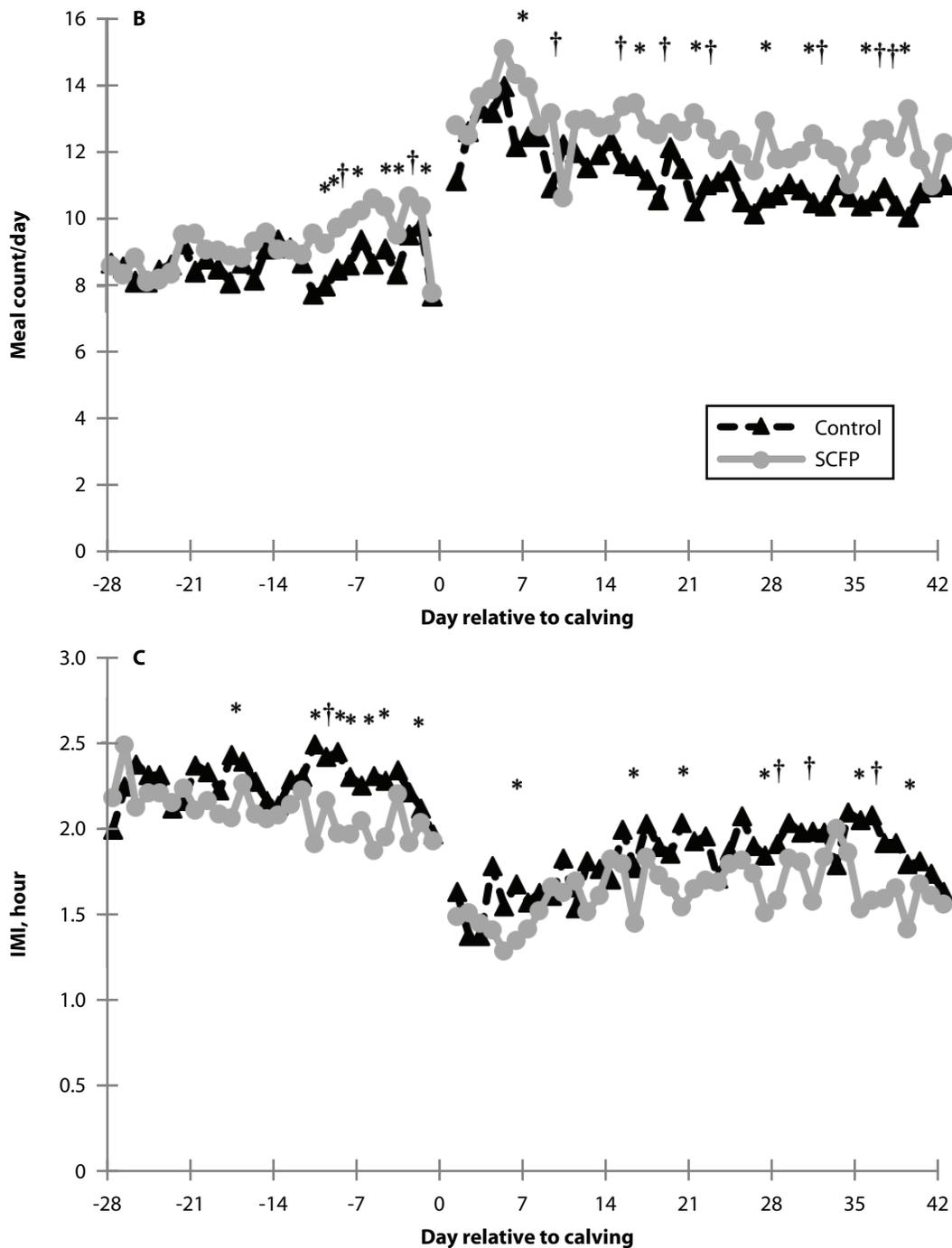
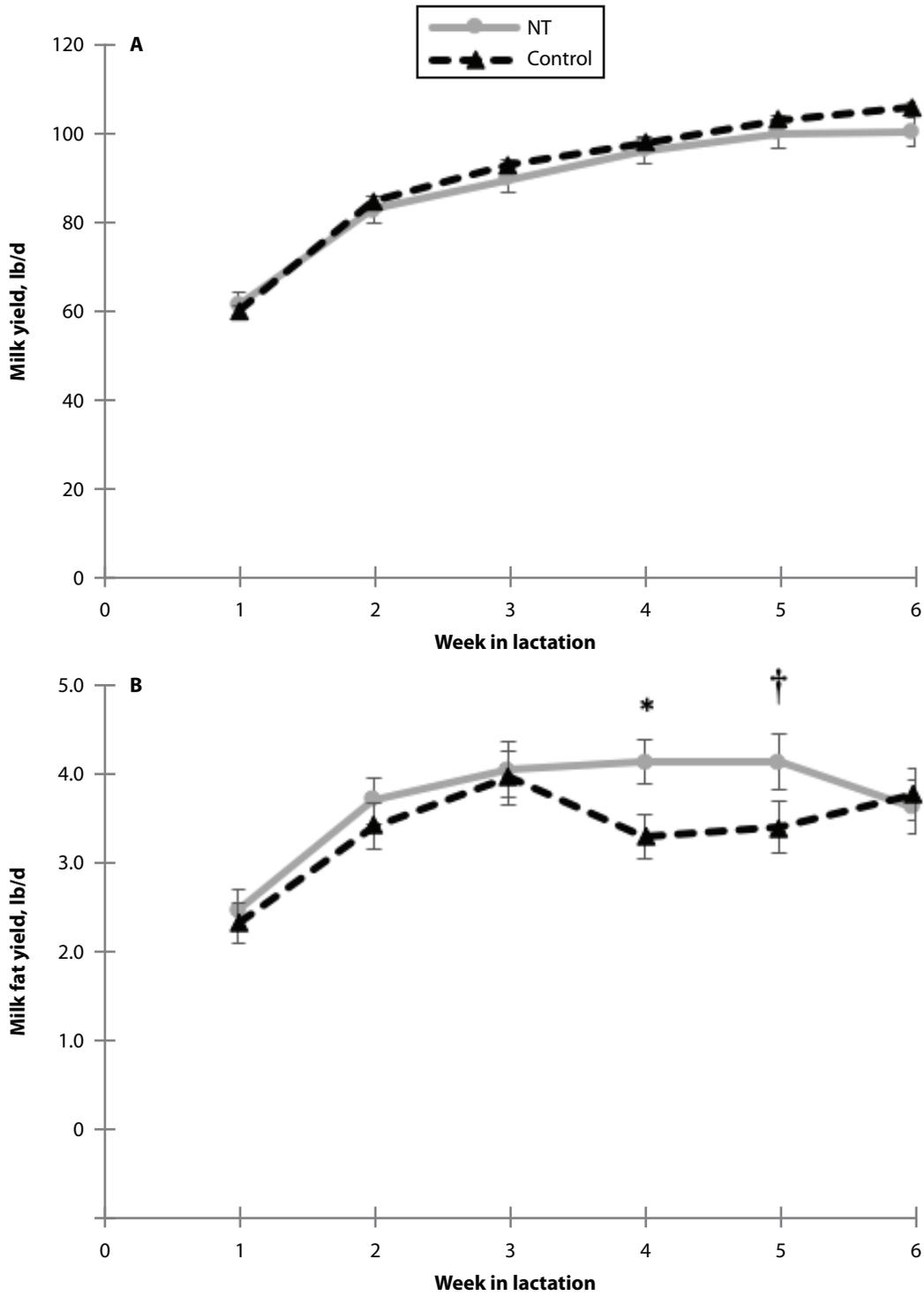


Figure 1. Dry matter intake (A), meal count (B), and inter-meal interval (IMI; C) for control cows and cows supplemented with *Saccharomyces cerevisiae* fermentation product (NutriTek, NT) from -29 ± 5 days relative to calving (DRTC) through 42 days in milk. An effect of time was present both prepartum and postpartum for all measures ($P < 0.001$). Treatment differences are indicated by * ($P < 0.05$) and † ($0.05 \leq P < 0.10$). A) DMI was not affected by NT ($P \geq 0.75$). B) NT cows tended to consume more meals per day prepartum ($P = 0.06$) and did increase meals per day postpartum ($P = 0.03$). Prepartum standard error of the means = 0.22, postpartum standard error of the means = 0.45. C) NT decreased time between meals prepartum ($P = 0.03$) and tended to decrease inter-meal interval postpartum ($P = 0.07$). Prepartum standard error of the means = 0.05, postpartum standard error of the means = 0.09.



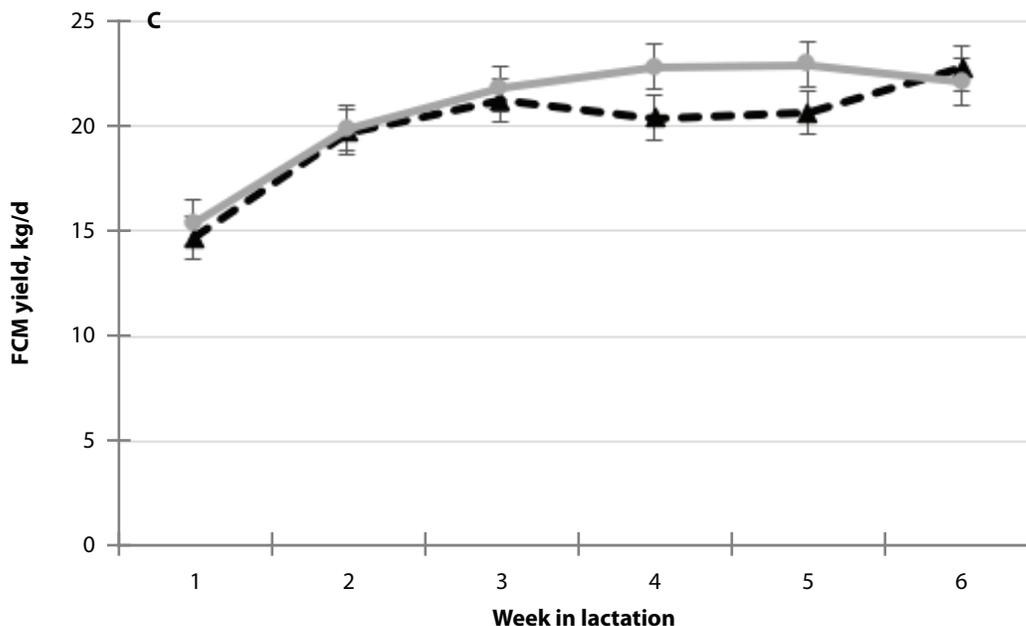


Figure 2. Milk yield, milk fat yield, and fat-corrected milk (FCM) of control cows and cows supplemented with *Saccharomyces cerevisiae* fermentation product (NutriTek, NT) from day -29 ± 5 relative to calving through 42 DIM. A) Milk yield did not differ between treatments ($P = 0.43$). There was an effect of week ($P < 0.001$), but no treatment \times week interaction ($P = 0.24$). B) Weekly milk fat yield was not different for cows supplemented with NT compared to control cows ($P = 0.10$). Milk fat yield differed by week ($P < 0.001$), and there was a tendency for a treatment \times week interaction ($P = 0.09$). Treatment differences are indicated by * ($P < 0.05$) and † ($0.05 \leq P < 0.10$). C) Fat-corrected milk did not differ between treatments ($P = 0.32$) and there was no treatment \times week interaction ($P = 0.20$).

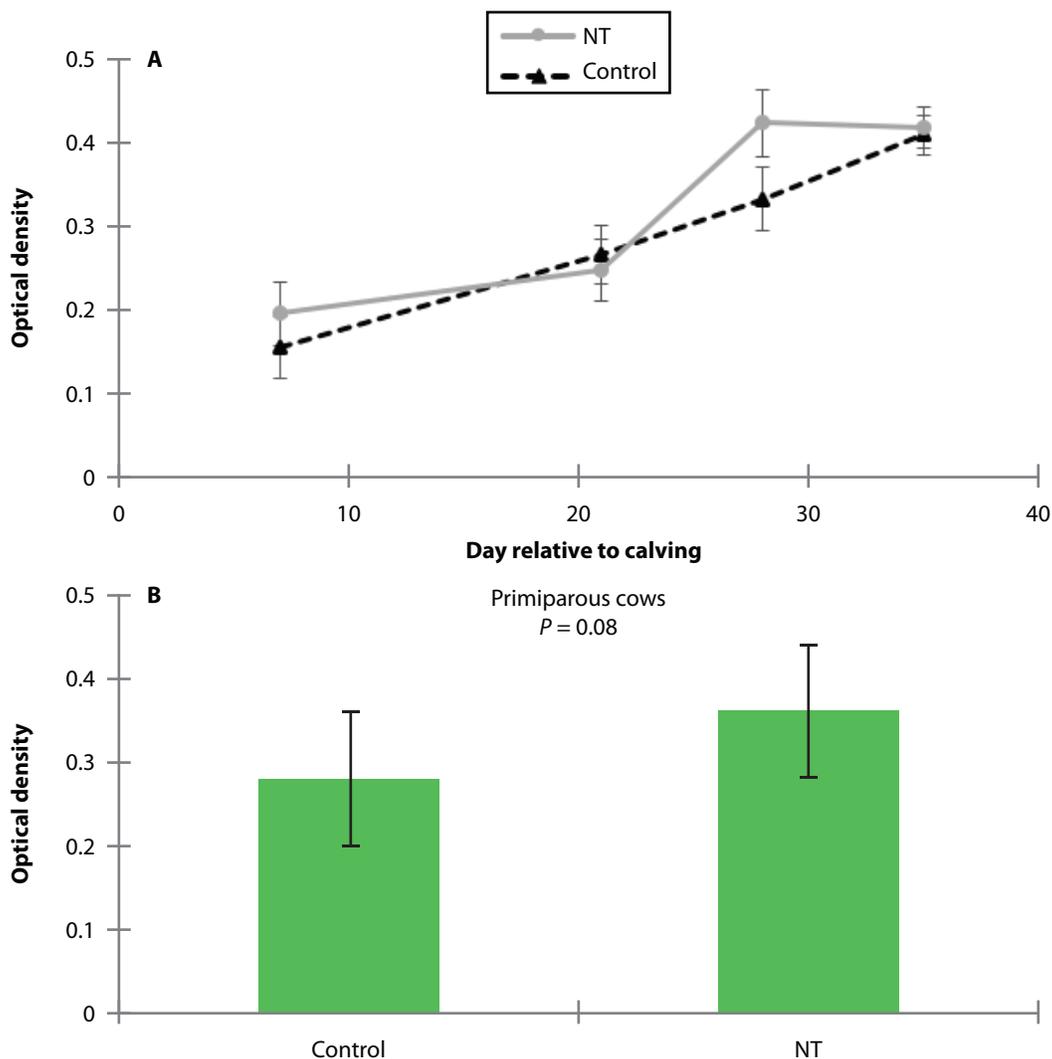


Figure 3. Serum concentrations of anti-ovalbumin-IgG in cows supplemented with *Saccharomyces cerevisiae* fermentation product (NutriTek, NT) from day -29 ± 5 relative to calving through 42 days in milk and challenged with a subcutaneous injection of ovalbumin on day 7 of lactation. **A)** Anti-ovalbumin IgG concentration increased with time after injection ($P < 0.001$); however, did not differ by treatment ($P = 0.25$). **B)** The tendency for a treatment \times parity interaction ($P = 0.08$) indicated greater anti-ovalbumin IgG concentration in primiparous NT cows compared to control.

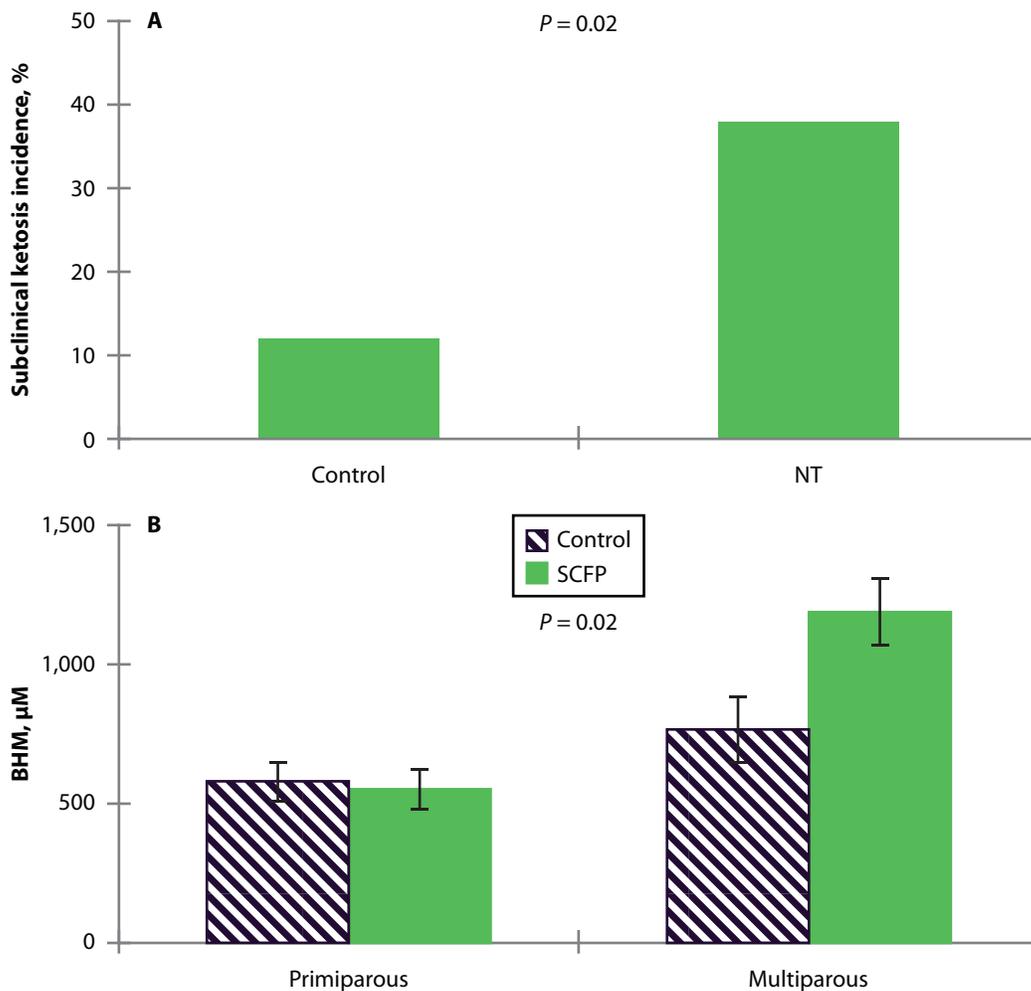


Figure 4. A) Supplementation with a *Saccharomyces cerevisiae* fermentation product (NT) from day -29 ± 5 relative to calving through 42 days in milk increased incidence of subclinical ketosis. B) When analyzed independently, week 2 β -hydroxybutyrate (BHB) concentrations demonstrated a significant parity \times treatment interaction with differences for multiparous but not primiparous cows.