Effects of wet corn gluten feed on ruminal pH and productivity of lactating dairy cattle fed diets with sufficient physically effective fiber

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ABSTRACT

Wet corn gluten feed (WCGF), a byproduct of the wet-milling industry, is commonly substituted in lactating dairy rations for both forages and concentrates. Previous research has shown that increasing WCGF in the diet decreased ruminal pH, likely due in part to decreasing particle size as forage inclusion rate decreased. The objective of this study was to maintain at least 10% of ration particles >19 mm in length across diets while increasing WCGF inclusion in the diet. We hypothesized that as WCGF increased in this scenario, dry matter intake (DMI) and milk yield would increase and ruminal pH would be maintained. Seven ruminally cannulated, lactating Holstein cows (4 multiparous and 3 primiparous) were used in an incomplete 4 × 4 Latin square design. Treatments included 0, 12.4, 24.5, or 35.1% WCGF and used alfalfa hay to maintain particle size. Across treatments, crude protein and neutral detergent fiber concentrations were held relatively constant. Four 21-d periods were used with 17 d of adaptation and 4 d of sample collection. Indwelling ruminal pH probes were used during sampling periods and recorded pH every 5 min. Particle size of total mixed rations andorts were analyzed using a Penn State Particle Separator (The Pennsylvania State University, University Park). Results were analyzed with mixed models to test the fixed effect of treatment. All diets contained ≥10% of particles >19 mm; however, as WCGF increased, the proportion of particles >19 mm decreased. Interestingly, with increasing WCGF, cows sorted for the particles >19 mm but against particles on the bottom screen and pan. With increasing WCGF, ruminal pH was not affected, but DMI and milk yield increased in a quadratic fashion, with the peak responses for the 24.5% WCGF diet. Milk protein, lactose, and fat concentrations were not affected by treatment; however, milk protein and lactose yields

increased with the inclusion of WCGF because of the increased milk yield. Production efficiency was not affected by treatments. Thus, if adequate particle size is maintained when WCGF increases in the diet, DMI and milk yield increase while maintaining production efficiency and ruminal pH.

Key words: ruminal pH, particle size, physically effective neutral detergent fiber

INTRODUCTION

As the demand for cereal grains increases, the use of alternative byproduct feeds becomes more appealing to dairy producers. One common byproduct of the corn wet-milling industry is wet corn gluten feed (WCGF). This is a high-fiber, highly digestible, moderate energy and CP feedstuff (Schroeder, 2003) that is commonly used to replace corn silage, corn grain, or alfalfa hay (or all of these) in lactating cow diets (Bernard and McNeill, 1991; Mullins et al., 2009). The study of Mullins et al. (2010) was designed to evaluate responses to WCGF replacing a combination of forages and concentrates in a lactation diet with minimal changes in dietary NDF, NFC, and CP concentrations. This study was particularly interesting because although increasing levels of WCGF increased DMI and milk production, it also decreased ruminal pH and efficiencies for milk production (Mullins et al., 2010). One reason for this may have been the small particle size of the WCGF, resulting in more rapid particulate passage rates, decreased saliva production, and decreased buffering in the rumen, thus decreasing ruminal pH. However, the role of particle size in these responses could not be evaluated in that particular study.

The importance of particle size in lactating cow diets has been summarized by Allen (1997) and is termed physically effective NDF (peNDF). This is a measure of a feedstuff's ability to stimulate chewing activity, which is the sum of time spent eating and ruminating (Beauchemin and Yang, 2005), often referred to as total chewing time. Studies have shown that increasing peNDF in lactating cow diets increases total chewing time, which results in increased saliva secretion due to

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mastication, and thus greater saliva flow into the rumen (Krause et al., 2002; Beauchemin and Yang, 2005). Saliva contains buffers such as bicarbonate and hydrogen phosphate that function to remove hydrogen ions from the rumen and, thus, aid in maintaining ruminal pH (Allen, 1997). Additionally, increases in particle size have been shown to increase acetate:propionate ratios (Zebeli et al., 2008), but this response depends heavily on the fermentability of diet ingredients (Krause and Combs, 2003; Beauchemin and Yang, 2005).

No studies have been performed that directly examine the effects of feeding increasing levels of WCGF while maintaining a minimum particle size measurement across treatment diets. The objective of this study was to evaluate the effects of WCGF inclusion (0, 12.4, 24.5, and 35.1% of diet DM) on ruminal pH, DMI, and milk production and efficiency while maintaining a minimum dietary particle size. We hypothesized that by maintaining at least 10% of particles >19 mm in all diets, ruminal pH and milk production efficiency would be consistent across all diets and feed intake and milk production would increase as WCGF inclusion rate increased.

MATERIALS AND METHODS

Experimental procedures were approved by the Institutional Animal Care and Use Committee at Kansas State University (Manhattan).

Design and Treatments

Four multiparous (2.8 ± 1.0 lactations; mean $\pm$ SD) and 3 primiparous cannulated, lactating Holstein cows (132 ± 22 DIM) were assigned to squares based on parity and randomly assigned within square to treatment sequence in a replicated incomplete 4×4 Latin square design to evaluate the effects of WCGF on production responses and ruminal pH. Initially, 8 cows were cannulated for the trial, but 1 was culled for health reasons before the trial. One multiparous cow was removed from the study after the completion of the second period due to health problems unrelated to treatments. The study consisted of 4 periods, each lasting a total of 21 d with 17 d of adaptation and 4 d for sample collection. To allow comparisons to be as direct as possible, diets were formulated exactly as they were for a previous study in which small ration particle size was thought to contribute to suppression of ruminal pH (Mullins et al., 2010). Diets were designed to include WCGF (Sweet Bran; Cargill Inc., Blair, NE) at 0, 12, 24, or 36% of diet DM and were formulated and mixed to have at least 10% of particles >19 mm. The purpose of fixing

the particle size was to maintain a minimum peNDF across all diets. Particle size goals were achieved by grinding alfalfa hay for 2 min in a horizontal mixing wagon (Roto-Mix, Dodge City, Kansas) before mixing the TMR in a stationary batch mixer. The formulations of the 4 diets, which were identical to those used in Mullins et al. (2010), are shown in Table 1, and the initial corn silage, alfalfa hay, and WCGF analyses are shown in Table 2. Although diets were designed to include 0, 12, 24, and 36% WCGF, actual concentrations fed were 0, 12.4, 24.5, and 35.1% (DM basis).

Cows were fed twice daily at 1100 and 1500 h to provide 110% of expected intake and were milked 3 times daily at 0200, 1000, and 1800 h. Animals were housed in a tie-stall barn with an evaporative cooling system from June through August 2010 at the Kansas State University Dairy Teaching and Research Facility.

Data and Sample Collection

Feed intakes were recorded daily as the difference between feed offered and refused. Throughout the treatment periods, corn silage and WCGF samples were analyzed twice weekly for DM and diets were adjusted accordingly. During sampling periods, individual ingredient samples as well as TMR and ort samples from each cow were collected daily. Water intakes were recorded daily during the sampling periods using in-line meters (model C700; Elster AMCO Water Inc., Ocala, FL). Body weight was measured and BCS assessed by 3 trained investigators on d 1 of each period and d 21 of the last period.

On d 18 through 21 of each period, milk samples were collected and yields were recorded at each milking. At 9-h increments on d 18 through 20, ruminal fluid samples for VFA and NH_3 determination were collected as grab samples from multiple locations in the ventral sac, composited, strained through 8 layers of cheese cloth, and stored at -20°C . Ruminal pH was recorded every 5 min during the sample period with indwelling ruminal pH probes (rumen sensors; Kahne Ltd., Auckland, New Zealand). Probes were calibrated and inserted into the rumen through the cannula on d 17, removed and recalibrated on d 19, and then placed back in the rumen until the completion of the sample period. Data from the first 24 h after each calibration (2 sets/period) were used for analysis. Probes recovered from the rumen were typically found in the ventral sac, most often in the central region and occasionally close to the reticulum. As such, the range of locations in which the probes were found was consistent with the sampling area from which ruminal VFA samples were collected.

Table 1. Ingredient and nutrient composition of dietary treatments

Item, % of diet DM	Inclusion of wet corn gluten feed (WCGF)			
	0%	12.4%	24.5%	35.1%
Ingredient				
Corn silage	19.9	23.0	19.8	16.3
WCGF	—	12.4	24.5	35.1
Alfalfa	24.5	24.8	21.1	17.4
Cottonseed	6.4	6.5	6.4	6.2
Ground corn grain	26.5	21.8	18.8	15.9
Soybean meal 48	8.6	4.9	2.2	2.2
Molasses	0.4	0.4	0.4	0.4
Soybean hulls	5.0	—	—	—
Limestone	1.0	1.1	1.3	1.4
Expeller soybean meal	3.3	3.7	4.0	3.6
Magnesium oxide	0.26	0.24	0.21	0.17
Micronutrient premix ¹	1.3	1.3	1.3	1.3
Nutrient				
DM, % as fed	63.1	60.3	60.3	60.8
CP	19.6	19.2	19.1	19.8
NDF	27.4	28.4	29.8	30.6
NDF provided by forage	18.5	18.7	16.0	13.2
peNDF >1.18 ²	23.9	25.7	27.1	28.2
Starch	27.6	26.3	24.5	22.5
NFC	41.9	41.5	40.5	39.3
Ether extract	4.4	4.4	4.4	4.3
Ash	8.2	8.3	8.4	8.4

¹Contained (DM basis): 1.2% Ca, 27.2% Na, 17.3% Cl, 93 mg/kg Co, 478 mg/kg Cu, 944 mg/kg Fe, 21 mg/kg I, 1,206 mg/kg Mn, 19 mg/kg Se, 2,145 mg/kg Zn, 307 IU/g vitamin A, 77 IU/g vitamin D, 1.11 IU/g vitamin E.

²Physically effective NDF (peNDF) calculated as dietary NDF × percentage of particles >1.18 mm (Mertens, 1997).

Feed and Milk Analyses

Milk samples were analyzed for fat, true protein, lactose (B-2000 Infrared Analyzer; Bentley Instruments Inc., Chaska, MN), MUN (MUN spectrophotometer; Bentley Instruments Inc.), and somatic cells (SCC 500; Bentley Instruments Inc.) by Heart of America DHIA (Manhattan, KS). Energy-corrected milk was calculated as follows: $0.327 \times \text{milk yield} + 12.86 \times \text{fat yield} + 7.65 \times \text{protein yield}$, and 3.5% FCM was calculated as $\text{milk yield} \times 0.432 + \text{fat yield} \times 16.216$ (Dairy Records Management Systems, 2010). For the evaluation of production efficiency, ECM was divided by DMI.

Samples of TMR and orts were evaluated for particle size using the 3-sieve Penn State Particle Separator (The Pennsylvania State University, University Park;

Kononoff et al., 2003). After analysis of individual ort and TMR samples, particle size distributions were averaged within cow for the 4-d sampling period and a sorting index was calculated using the procedure outlined by Silveira et al. (2007). In this index, a value of 1.0 indicates that no sorting occurred, whereas values <1.0 indicate that animals sorted against the fraction, and values >1.0 indicate that animals selected for the fraction. Separated samples as well as ingredient samples were dried in a 55°C forced-air oven for 72 h and DM was determined. Samples were ground to pass through a 1-mm screen in a Wiley Mill (Arthur H. Thomas Co., Philadelphia, PA) and then composited by period. True DM was determined by drying samples at 105°C in a forced-air oven for 24 h; nutrient concentrations are reported relative to this value. The concentration of ash

Table 2. Composition of corn silage, alfalfa hay, and wet corn gluten feed (WCGF)

Nutrient, % of diet DM	Corn silage	Alfalfa hay	WCGF
DM (55°C), % as fed	35.1	90.5	64.6
DM (105°C), % as fed	33.0	84.7	59.4
NDF	37.3	40.7	37.9
CP	8.0	25.0	22.5
Starch	36.7	3.2	13.5
Ether extract	3.6	2.4	2.1
Ash	5.5	9.9	5.5

was determined by oxidizing samples at 500°C for 8 h in a muffle furnace. Neutral detergent fiber concentrations were determined (Van Soest et al., 1991) in the presence of α -amylase and sodium sulfite with an Ankom fiber analyzer (Ankom Technology, Fairport, NY). Starch content was determined by digesting samples in α -amylase and glucoamylase and then determining glucose quantities with a commercial kit (Autokit Glucose; Wako Chemicals USA Inc., Richmond, VA). Crude protein concentration was determined by oxidation and detection of N_2 (Leco analyzer; Leco Corp., St. Joseph, MI). Ether extract was used to determine crude fat concentrations (AOAC, 2000; method 920.9).

Ruminal VFA and NH_3 Analysis

Ruminal fluid samples were thawed and composited by cow and period to obtain an 8-mL sample. Samples were then mixed with 2 mL of 25% metaphosphoric acid and were centrifuged at $30,000 \times g$ for 20 min at 24°C. The supernatant was collected for VFA and NH_3 determination. Ammonia was determined colorimetrically with an autoanalyzer (Technicon Analyzer II; Technicon Industrial Systems, Buffalo Grove, IL). Ruminal VFA were measured with a gas chromatograph (model 5890; Hewlett-Packard, Avondale, PA) with a flame ionization detector. The chromatograph was fitted with a 1.82×6.35 -mm-i.d. glass column packed with GP 10% SP-1200/1% H_3PO_4 (Supelco #1-1965; Supelco Inc., Bellefonte, PA). The column was maintained at 130°C, the detector and injector were maintained at 250°C, and the carrier gas (N_2) flow was 80 mL/min. The run time was approximately 5 min.

Statistical Analysis

Data were analyzed by JMP 8.0 (SAS Institute Inc., Cary, NC), with cow and period as random effects.

Square, treatment, and square by treatment interaction were included as fixed effects. When the P -value for the square by treatment interaction was >0.6 , the term was removed from the model. Responses to WCGF inclusion rate were tested with linear and quadratic contrasts, which were declared significant at $P < 0.05$, and tendencies were identified if $0.05 < P < 0.10$.

RESULTS

Particle Size

Although all diets contained at least 10% of particles on the top screen, a linear decrease occurred in particles >19 mm as WCGF increased in the diet (Table 3). Conversely, a linear increase was found in particles on the bottom screen (1.18 – 8 mm) as WCGF increased. This result can be explained by the reduction in corn silage and alfalfa as WCGF was increased in the diet, resulting in a decrease in the number of large particles. The sorting index analysis (Table 3) revealed that as WCGF increased in the ration, selection for larger particles increased in a quadratic fashion, whereas selection of particles on the bottom screen (1.18 – 8 mm) and pan (<1.18 mm) linearly decreased.

Feed Intake and Milk Production

Intake and production variables are presented in Table 4. A quadratic effect on DMI was observed, with the highest intake for the 24.5% WCGF diet. Milk production responded similarly to intake, with a quadratic increase, and the 24.5% WCGF treatment again yielded the greatest response, with production of 41.4 ± 3.6 kg/d (mean $\pm$ SEM). No differences were observed in 3.5% FCM, BCS, or BW between diets; however, ECM yield tended to increase linearly with in-

Table 3. Particle size distribution and sorting index

Item	Inclusion of wet corn gluten feed (WCGF)				SEM	P -value	
	0%	12.4%	24.5%	35.1%		Linear	Quadratic
TMR, ¹ % of DM							
>19 mm	15.0	13.6	12.3	10.1	1.88	0.01	0.71
8–19 mm	25.8	27.3	30.6	27.2	1.95	0.45	0.24
1.18–8 mm	47.0	49.4	48.0	54.2	1.50	0.01	0.28
<1.18 mm	12.6	9.5	8.9	7.8	1.23	<0.01	0.01
Sorting index ²							
>19 mm	0.95	0.90	0.95	1.04	0.04	0.04	0.04
8–19 mm	0.99	0.99	0.99	1.01	<0.01	0.27	0.35
1.18–8 mm	1.01	1.02	1.00	0.98	0.01	<0.01	0.09
<1.18 mm	1.04	1.03	1.02	1.00	0.01	0.01	0.75

¹Values are means across all 4 periods, $n = 16$ per treatment.

²Calculated from methods outlined by Silveira et al. (2007): >1.0 = sorting in favor, <1.0 = sorting against. Mean refusals as percentage of feed offered: 0% WCGF = 14.6%, 12.4% WCGF = 13.6%, 24.5% WCGF = 12.4%, and 35.1% WCGF = 16.0%.

Table 4. Effects of treatments on intake and performance of lactating cows

Item	Inclusion of wet corn gluten feed				SEM	<i>P</i> -value	
	0%	12.4%	24.5%	35.1%		Linear	Quadratic
DMI, kg/d	25.2	26.6	27.0	26.5	1.17	0.02	0.02
Water intake, L/d	129	129	140	139	7.9	0.11	0.88
Milk, kg/d	37.7	40.8	41.4	40.7	3.64	0.01	0.02
3.5% FCM, kg/d	34.8	37.3	36.7	36.8	3.06	0.19	0.18
ECM, kg/d	35.3	37.9	37.6	37.7	3.03	0.08	0.14
ECM/DMI	1.39	1.43	1.39	1.42	0.08	0.60	0.92
BW change, kg/21 d	-2.7	-11.1	-13.8	-6.3	8.80	0.58	0.14
BCS change/21 d	-0.10	-0.05	-0.08	0.03	0.05	0.14	0.56

creasing levels of WCGF. When evaluating production efficiency (ECM/DMI), no differences were detected between treatments.

Across all treatments, milk fat concentration, lactose concentration, and fat yield were similar (Table 5); however, milk protein concentration tended to increase quadratically ($P = 0.09$) as WCGF inclusion rates increased, and a quadratic response was observed for MUN. Furthermore, milk protein yield increased linearly with WCGF inclusion and milk lactose yield showed a quadratic response. The quadratic increase in milk lactose yield was primarily a result of the increase in milk production because treatments did not alter lactose concentration (Table 5).

Ruminal pH and VFA

Mean ruminal pH ranged from 6.00 to 6.13 across treatments (Table 6); neither time nor area under pH 5.8 or 5.6 were affected by treatment. Total ruminal VFA concentration was not affected by treatment (Table 7); however, acetate concentration linearly decreased with increasing levels of WCGF. Propionate, butyrate, and lactate concentrations were not affected by WCGF inclusion level. As WCGF increased in the diet, isobutyrate and isovalerate concentrations linearly decreased, whereas valerate concentrations linearly increased. Although propionate concentration was not

affected, the decrease in acetate levels was great enough to lead to a decrease in the acetate:propionate ratio.

DISCUSSION

The present study was conceived as a relatively direct comparison to previous work (Mullins et al., 2010) in which the ration formulation was identical (with very similar forage sources) but with the intent of feeding a diet with substantially greater particle size. In both studies, dietary starch content decreased with increasing WCGF. In the present experiment, NDF concentrations were slightly lower across all treatments, but followed similar trends as those fed by Mullins et al. (2010). The most dramatic difference between the studies was the particle size distribution of all diets. The present study maintained consistently higher levels of particles >19 mm, with ranges of 10.1 to 15.0% compared with 2.36 to 3.85% in the previous study (Mullins et al., 2010). Another large difference between the 2 trials was the proportion of particles <8 mm, with the present study providing fewer particles below this level than did Mullins et al. (2010). These differences in particle size may play a crucial role in the differences seen in intake, yields, efficiency, and ruminal pH (Yang and Beauchemin, 2006).

Dietary NDF concentrations were relatively low in this study. The NRC (2001) provided recommendations

Table 5. Effects of treatments on milk components

Item	Inclusion of wet corn gluten feed				SEM	<i>P</i> -value	
	0%	12.4%	24.5%	35.1%		Linear	Quadratic
Milk fat, %	3.05	2.99	2.83	2.97	0.16	0.24	0.17
Milk protein, %	2.94	2.92	2.93	3.01	0.12	0.10	0.09
Lactose, %	4.84	4.87	4.86	4.84	0.07	0.93	0.29
MUN, mg/dL	13.2	12.6	12.9	13.6	0.82	0.30	0.04
Yield, kg/d							
Milk fat	1.14	1.21	1.16	1.18	0.11	0.73	0.53
Milk protein	1.09	1.18	1.20	1.21	0.09	<0.01	0.09
Milk lactose	1.82	1.98	1.99	1.96	0.16	0.03	0.02

Table 6. Effects of treatments on rumen pH

Item	Inclusion of wet corn gluten feed				SEM	P-value	
	0%	12.4%	24.5%	35.1%		Linear	Quadratic
pH	6.05	6.13	6.00	6.00	0.07	0.26	0.45
SD	0.42	0.40	0.40	0.44	0.02	0.58	0.19
Time under 5.8, min/d	434	304	467	465	72.9	0.31	0.24
Area under 5.8, pH $\times$ min/d	130	88	144	138	28.5	0.50	0.46
Time under 5.6, min/d	257	175	290	293	56.4	0.33	0.39
Area under 5.6, pH $\times$ min/d	62	39	73	61	16.6	0.65	0.74

for NDF as a function of NDF derived from forage sources, because forage sources of NDF are more effective in maintaining milk components. For example, the minimum NDF recommended for a diet containing 19% NDF from forage is 25%, whereas a diet containing only 15% NDF from forage requires at least 33% NDF (NRC, 2001). In the 0 and 12.4% diets, NDF requirements were satisfactory in terms of requirements outlined by the NRC; however, the 24.5 and 35.1% diets are both deficient in NDF according to these standards, because the NRC would require a minimum of 31 and >33% NDF for these levels of forage NDF. The lack of a negative effect on performance from these 2 diets, especially in light of the decrease in efficiency observed in the previous study (Mullins et al., 2010), highlights the important role of particle size in the present study.

Mullins et al. (2010) and the current study showed similar responses in multiple production variables, demonstrating increases in DMI, milk yield, ECM, milk protein yield, and milk lactose yield as WCGF inclusion rates increased. These results parallel those of VanBaale et al. (2001), who fed 0 or 20% WCGF to cows averaging 147 ± 55 DIM (mean $\pm$ SD). These results differ largely from those of Schroeder (2003), who found no differences in DMI, milk yield, milk fat, and milk protein concentrations, or efficiencies when feeding WCGF at levels between 0 and 45% DM in a corn silage and alfalfa haylage diet. One very clear difference

between diets used in the present study compared with those of Schroeder (2003) was the carbohydrate profile; our diets included approximately 29% NDF and 41% NFC, whereas Schroeder (2003) fed diets with approximately 43% NDF and 25% NFC. Nonforage NDF from WCGF replaced both forage NDF and starch in each of these studies, but the linear increases in DMI and milk production in the present study may have been a response to decreases in dietary starch and the negative associative effects on NDF digestion. This putative improvement in ruminal NDF digestion as starch was removed would have been less likely to occur in the lower-NFC diets fed by Schroeder (2003).

In the study by Mullins et al. (2010), milk fat yield increased but milk protein concentration did not change as WCGF was added. This result differs from the present study, where milk fat yields were not statistically different between diets and milk protein concentration tended to increase quadratically. Other differences occurred between the 2 studies, including a decrease in efficiency and an increase in BCS in the study by Mullins et al. (2010), suggesting that energy was directed toward adipose accretion in diets containing high levels of WCGF. In the present study, efficiencies were maintained across all treatments along with BCS, which suggests that energy consumed in the present study was partitioned in a consistent manner across diets. One possible reason for this observed difference

Table 7. Effects of treatments on ruminal VFA

Item	Inclusion of wet corn gluten feed				SEM	P-value	
	0%	12.4%	24.5%	35.1%		Linear	Quadratic
Total VFA, mM	129.3	135.7	128.1	123.1	5.65	0.20	0.20
Acetate, mM	77.4	78.3	71.9	66.8	2.83	<0.01	0.26
Propionate, mM	31.2	35.1	34.6	33.9	2.45	0.18	0.08
Butyrate, mM	14.8	15.7	15.3	15.8	0.71	0.37	0.75
Isobutyrate, mM	1.04	1.02	0.91	0.86	0.05	<0.01	0.78
Isovalerate, mM	0.91	0.86	0.74	0.62	0.09	<0.01	0.58
Lactate, mM	0.18	0.91	0.87	0.51	0.33	0.57	0.15
Valerate, mM	2.58	3.08	3.23	3.68	0.24	<0.01	0.92
Acetate:propionate	2.58	2.26	2.08	2.00	0.11	<0.01	0.03
NH ₃ , mM	6.44	7.66	7.23	6.81	0.57	0.76	0.10

in the partitioning of energy could be the result of the proportions of acetate and propionate. Diets that promote increased production of propionate can increase insulin concentrations because of propionate's ability to stimulate insulin secretion (Bradford et al., 2006). Insulin, in turn, is a key adipogenic hormone that may promote repartitioning of energy to adipose tissue (Voelker and Allen, 2003; Mahjoubi et al., 2009). In the present study, only a trend was detected for a quadratic increase in propionate, whereas Mullins et al. (2010) reported an increase in propionate concentrations with increasing WCGF.

Another area of interest between these studies is the effect of WCGF and particle size on ruminal measures. In the study by Mullins et al. (2010), ruminal pH decreased with increasing WCGF; however, in the present study, no statistical differences were observed in ruminal pH between treatments. Despite the changes across treatments in particles >19 mm in the present study, the relative abundance of large particles in all treatments may help to explain the consistency of ruminal pH by stimulating chewing activity, increasing salivary secretions, and ultimately increasing ruminal buffering. Although particles >19 mm in length decreased with increasing WCGF, peNDF actually increased due to increasing NDF of the diets. Another possible explanation could be that the large particles slowed passage rates and, thus, decreased feed intake and the rapid fermentation that can occur after large meals. Although DMI was slightly lower in the present study than in Mullins et al. (2010), it is difficult to compare these values to determine if this difference was enough to affect ruminal pH.

Under extreme conditions such as SARA, modifications in ruminal fermentation can arise and often lead to milk fat depression and depressed DMI (Kleen et al., 2003). Kleen et al. (2003) defined SARA as present when mean ruminal pH is ≤ 5.5 and absent when mean ruminal pH is ≥ 5.8 . Zebeli et al. (2008), who conducted a meta-analysis of 100 studies, concluded that mean ruminal pH should not decrease below 6.16 and that time under pH 5.8 should not exceed 5.24 h/d to minimize the risk of SARA. Based on these criteria and using mean pH as the defining feature of SARA, we found no evidence that SARA was present in any of the treatment diets, although time under pH 5.8 was greater than 5.24 h/d for all treatment diets except the 12.4% WCGF diet, which may help explain the low milk fat concentrations in this study. Krause and Oetzel (2006) determined that pH can range from 0.5 to 1.0 pH units around the mean and, along with diurnal pH fluctuations, mean pH values should be used with caution (Palmonari et al., 2010). In this study, pH data

were collected continuously, which adds confidence to the mean pH values reported.

Another interesting observation from the present study was the finding that as particle size decreased, cows selected for larger particles. This increased selection for large particles, which may aid in maintaining ruminal pH and function, is consistent with the theory presented by Cooper et al. (1996), which states that when given the opportunity, ruminants will sort in favor of particles that aid in maintaining a healthy rumen environment. These results are similar to those of DeVries et al. (2008), who used mid lactation (low-risk) and early lactation (high-risk) cows to evaluate the effects of acidosis on sorting. When cows were not challenged with acidosis, they tended to sort for medium-length particles and against the longest and shortest particles. When high-risk cows were challenged with acidosis, they demonstrated the greatest degree of sorting and sorted in favor of longer particles (DeVries et al., 2008). Beauchemin and Yang (2005) found similar patterns in sorting for longer particles when cows had low ruminal pH. Although multiple studies have demonstrated this behavior, the level of ruminal pH at which this behavior is observed is not known. The physiological sensory mechanisms that initiate this selection of long particles and the learning process that may contribute to such behavior remain poorly understood (Keunen et al., 2002).

CONCLUSIONS

When WCGF was increased to 24.5% of diet DM, DMI, milk yield, and protein yield were increased and ruminal pH and production efficiencies were maintained, likely in part because of adequate particle size. The study was successful in maintaining at least 10% of the diet as particles >19 mm, although a linear decrease in particles >19 mm was found as WCGF inclusion increased. The quadratic responses seen in this study suggest that, although milk protein yield increased linearly, including WCGF at 24.5% of diet DM resulted in the most desirable production responses while maintaining consistent ruminal pH. Additionally, results from this study support the theory that ruminants, when given the opportunity, will sort for particles that help maintain a healthy ruminal environment.

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REFERENCES

- Allen, M. S. 1997. Relationship between fermentation acid production in the rumen and the requirement for physically effective fiber. *J. Dairy Sci.* 80:1447–1462.
- AOAC (Association of Official Analytical Chemists). 2000. *Official Methods of Analysis*. 17th ed. AOAC, Arlington, VA.
- Beauchemin, K. A., and W. Z. Yang. 2005. Effects of physically effective fiber on intake, chewing activity, and ruminal acidosis for dairy cows fed diets based on corn silage. *J. Dairy Sci.* 88:2117–2129.
- Bernard, J. K., and W. W. McNeill. 1991. Effect of high fiber energy supplements on nutrient digestibility and milk production of lactating dairy cows. *J. Dairy Sci.* 74:991–995.
- Bradford, B. J., M. Oba, R. A. Ehrhardt, Y. R. Boisclair, and M. S. Allen. 2006. Propionate is not an important regulator of plasma leptin concentration in dairy cattle. *Domest. Anim. Endocrinol.* 30:65–75.
- Cooper, S. D., I. Kyriazakis, and J. D. Oldham. 1996. The effects of physical form of feed, carbohydrate source, and inclusion of sodium bicarbonate on the diet selections of sheep. *J. Anim. Sci.* 74:1240–1251.
- Dairy Records Management Systems. 2010. DHI glossary. Accessed Sep. 15, 2010. <http://www.drms.org/PDF/materials/glossary.pdf>.
- DeVries, T. J., F. Dohme, and K. A. Beauchemin. 2008. Repeated ruminal acidosis challenges in lactating dairy cows at high and low risk for developing acidosis: Feed sorting. *J. Dairy Sci.* 91:3958–3967.
- Keunen, J. E., J. C. Plaizier, L. Kyriazakis, T. F. Duffield, T. M. Widowski, M. I. Lindinger, and B. W. McBride. 2002. Effects of a subacute ruminal acidosis model on the diet selection of dairy cows. *J. Dairy Sci.* 85:3304–3313.
- Kleen, J. L., G. A. Hooijer, J. Rehage, and J. P. T. M. Noordhuizen. 2003. Subacute ruminal acidosis (SARA): A review. *J. Vet. Med. A Physiol. Pathol. Clin. Med.* 50:406–414.
- Kononoff, P. J., A. J. Heinrichs, and D. R. Buckmaster. 2003. Modification of the Penn State forage and total mixed ration particle separator and the effects of moisture content on its measurements. *J. Dairy Sci.* 86:1858–1863.
- Krause, K. M., and D. K. Combs. 2003. Effects of forage particle size, forage source, and grain fermentability on performance and ruminal pH in midlactation cows. *J. Dairy Sci.* 86:1382–1397.
- Krause, K. M., D. K. Combs, and K. A. Beauchemin. 2002. Effects of forage particle size and grain fermentability in midlactation cows II. Ruminal pH and chewing activity. *J. Dairy Sci.* 85:1947–1957.
- Krause, K. M., and G. R. Oetzel. 2006. Understanding and preventing subacute ruminal acidosis in dairy herds: A review. *Anim. Feed Sci. Technol.* 126:215–236.
- Mahjoubi, E., H. Amanlou, D. Zahmatkesh, M. Ghelich Khan, and N. Aghaziarati. 2009. Use of beet pulp as a replacement for barley grain to manage body condition score in over-conditioned late lactation cows. *Anim. Feed Sci. Technol.* 153:60–67.
- Mertens, D. R. 1997. Creating a system for meeting the fiber requirements of dairy cattle. *J. Dairy Sci.* 80:1463–1481.
- Mullins, C. R., K. N. Grigsby, D. E. Anderson, E. C. Titgemeyer, and B. J. Bradford. 2010. Effects of feeding increasing levels of wet corn gluten feed on production and ruminal fermentation in lactating dairy cows. *J. Dairy Sci.* 93:5329–5337.
- Mullins, C. R., K. N. Grigsby, and B. J. Bradford. 2009. Effects of alfalfa hay inclusion rate on productivity of lactating dairy cattle fed wet corn gluten feed-based diets. *J. Dairy Sci.* 92:3510–3516.
- NRC. 2001. *Nutrient Requirements of Dairy Cattle*. 7th rev. ed. Natl. Acad. Press, Washington, DC.
- Palmonari, A., D. M. Stevenson, D. R. Mertens, C. W. Cruywagen, and P. J. Weimer. 2010. pH dynamics and bacterial community composition in the rumen of lactating dairy cattle. *J. Dairy Sci.* 93:279–287.
- Schroeder, J. W. 2003. Optimizing the level of wet corn gluten feed in the diet of lactating dairy cows. *J. Dairy Sci.* 86:844–851.
- Silveira, C., M. Oba, W. Z. Yang, and K. A. Beauchemin. 2007. Selection of barley grain affects ruminal fermentation, starch digestibility and productivity of lactating dairy cows. *J. Dairy Sci.* 90:2860–2869.
- Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber, non-starch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74:3583–3597.
- VanBaale, M. J., J. E. Shirley, E. C. Titgemeyer, A. F. Park, M. J. Meyer, R. U. Lindquist, and R. T. Ethington. 2001. Evaluation of wet corn gluten feed in diets for lactating dairy cows. *J. Dairy Sci.* 84:2478–2485.
- Voelker, J. A., and M. S. Allen. 2003. Pelleted beet pulp substituted for high-moisture corn: 1. Effects on feed intake, chewing behavior, and milk production of lactating dairy cows. *J. Dairy Sci.* 86:3542–3552.
- Yang, W. Z., and K. A. Beauchemin. 2006. Physically effective fiber: Method of determination and effects on chewing, ruminal acidosis, and digestion by dairy cows. *J. Dairy Sci.* 89:2618–2633.
- Zebeli, Q., J. Dijkstra, M. Tafaj, H. Steingass, B. N. Ametaj, and W. Drochner. 2008. Modeling the adequacy of dietary fiber in dairy cows based on the responses of ruminal pH and milk fat production to composition of the diet. *J. Dairy Sci.* 91:2046–2066.