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EFFECT OF SUBACUTE RUMINAL ACIDOSIS ON TMR PREFERENCE IN LACTATING DAIRY COWS

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ABSTRACT

Subacute ruminal acidosis (SARA) is a condition where the pH of the rumen becomes abnormally acidic, because of increased and altered production of volatile fatty acids (**VFA**). A common cause of acidosis is overeating large amounts of readily available nutrients, specifically carbohydrates. Acidic by-products of fermentation are responsible for the change in the rumen environment. Salivary bicarbonate neutralizes acids during normal rumination, which is initiated by forage, specifically long fiber particles. Ruminal acidosis can lead to cows going off-feed, compromising the health of the animal. In this study, 8 multiparous rumen-cannulated Holstein dairy cows were each given a choice between a long, slow-fermenting and a short, fast-fermenting ration. Feed intake was monitored beginning after adaptation on d 8. Rumen parameters (pH, VFA concentration) were monitored beginning d 11. On d 12, cows were restricted to 75% ad libitum intake; rumen environments were monitored to demonstrate changes created by the feed restriction. Following this, on d 13 cows were given 4 kg wheat grain followed by ad libitum long and short rations. Rumen pH was decreased by the grain feeding, and was accompanied by an increase in percent of total DMI consumed as long and slow fermenting TMR. The cows can increase their intake of long particle size and slower fermenting grain when experiencing this bout of SARA.

TABLE OF CONTENTS

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1. Introduction

1.1. Background

As the dairy industry moves forward, cows are being pushed to produce increasingly large amounts of milk. This increases dietary energy requirements for lactating cows, which in turn increases the need to stabilize the rumen environment and therefore for physically effective NDF (**peNDF**). Cows require fiber of adequate length to support the rumen mat, stimulate chewing, and thereby buffer the rumen. If dietary forage particles are too short, rumen buffering will not occur sufficiently and cows will become susceptible to subacute ruminal acidosis (**SARA**). If dietary forage particles are created too long in an attempt to overcome this SARA, this will result in reduced feed intake for high producing cows (Kononoff et al., 2003b). Because of these conflicting factors, it is difficult to describe the most appropriate forage particle size distribution for dairy cow TMR.

Despite low incidence rates of acute acidosis, the prevalence of SARA on commercial dairies in the U.S. has been estimated to be greater than 20 % (Krause and Oetzel, 2005; Dohme et al., 2008). It is important to understand the connection between diet and rumen health so that the detrimental effects associated with SARA can be avoided.

1.2. Fiber Requirements for Ruminants

The National Research Council (2001) recommends formulating lactating cow diets with a minimum NDF level of 25% of dietary DM, provided that alfalfa or corn silage are used as the predominant forage, dry ground corn grain is used as the predominant source of starch, and that 19% of DM NDF comes from forage. The specific conditions upon which these fiber recommendations are based make it difficult to apply them to many situations. The NRC (2001) suggests increasing minimum NDF levels and decreasing maximum NSC levels as forage NDF decreases or when diets are finely chopped. Minimum NDF levels should also be increased if dry corn grain is replaced with a more readily fermentable carbohydrate source.

In addition to basic chemical NDF requirements, cows also require fiber of adequate length for proper rumen function. The rumen mat, composed of longer forage particles, regulates ruminal fermentation, passage, and rumination (Mertens, 1997). Chewing activity initiates salivary $NaHCO_3$ secretion (Krause et al., 2002), which helps maintain a constant rumen environment and consequently provides for more consistent microbial fermentation patterns (Mertens, 1997). Forage particle length and NDF content do not affect salivation rate, but influence the amount of saliva produced by affecting eating rate and time spent eating (Beauchemin et al., 2008). It is important not to include too much effective fiber in dairy cow rations, however. Diets too high in peNDF may reduce feed intake, reduce feed efficiency, and negatively impact milk production by increasing the passage rate of potentially ruminally degradable starch from the rumen (Zebeli et al., 2010).

1.3. Measuring Physical Fiber

Mertens (1997) has described effective NDF (**eNDF**) as a measure of the ability of a feed to replace forage in a ration and maintain milk fat percentage. Effectiveness factors range from 0, where a feed has no ability to maintain milk fat percentage, to greater than 1.0, where it maintains milk fat percentage entirely. Because milk fat content is occasionally insensitive to dietary changes, peNDF is more commonly used as an indicator of fiber effectiveness (Mertens, 1997). In addition, peNDF is a more sensitive indicator than eNDF for the prevention of SARA, lameness, displaced abomasum, or intake depression (Mertens, 1997), and is therefore more commonly used to predict rumen health.

Mertens (1997) has described peNDF as the NDF concentration of the ration multiplied by the percent of particles retained on a 1.18 mm sieve. This definition relies on the assumptions that NDF is distributed uniformly over all particle sizes, that all particles larger than 1.18 mm create equal chewing times, and that the fragility, or ease of particle size reduction, is not different among NDF sources in the ration. The 1.18 mm sieve is used because of the importance

of fiber length in the formation of the rumen mat. Particles less than 1.2 mm in length are believed to pass readily out of the rumen (Mertens, 1997).

Many studies have used the 1.18 mm sieve in the Penn State Particle Separator (**PSPS**) to determine peNDF content of a forage or TMR (Yang and Beauchemin, 2006; Yang and Beauchemin, 2007; Bhandari et al., 2008), whereas others have used only the proportion retained in the 8 mm screen (Plaizier, 2004; DeVries et al., 2007) or the 19 mm screen (Krause et al., 2002). Therefore, there is currently some discrepancy in the literature as to what is the best measure for peNDF. Mertens (1997) used vertical shaking to separate particles, and observed that separation was based on their smallest dimension – cross-sectional width. The PSPS uses horizontal shaking to separate particles (Lammers et al., 1996), where separation is based on length. peNDF values calculated based on different particle size determination should not be considered equal substitutes.

1.4. Factors Affected by peNDF

Finely chopped forage often has increased digestibility because of extra surface area available for microbial fermentation (Yang and Beauchemin, 2006). This may allow for an increase in total rumen VFA concentration (Kononoff et al., 2003b). Increases in digestibility are beneficial to the cow if the rate of passage is sufficiently slow enough to allow optimal time for fermentation and digestion of the feed. Conversely, increasing forage particle size at some point decreases digestibility and total VFA concentration (Krause et al., 2002). Increased fiber concentration decreases total VFA production and increases chewing activity per kg of fiber ingested (Mertens, 1997).

Total chewing time is a good measure of a feed's physical effectiveness (Sudweeks et al., 1981). Increasing ration particle size decreases DMI (Krause et al., 2002; Maulfair et al., 2010) and increases chewing activity (Maulfair et al., 2010). Conversely, as particle size is reduced, chewing activity per kilogram of DMI decreases linearly (Kononoff et al., 2003b). DMI from the

pan of the PSPS has been shown to be negatively correlated to time spent ruminating, while DMI from the top screen is positively correlated with ruminating time (Krause et al., 2002). Decreased chewing activity leads to less salivary buffering of the rumen, which allows for a lower overall rumen pH. Low acetate:propionate ratios have been observed as a result of this alteration, which may be responsible for milk fat depression (Mertens, 1997).

Rumen pH may be a better indicator of rumen health than milk fat content (Mertens, 1997). Krause et al. (2002) reported that milk fat content was not correlated to chewing or intake, but instead to mean rumen pH and time spent below pH 5.8. Diets higher in peNDF result in increases in rumen pH and fewer pH fluctuations throughout the day (Stone, 2004), which provide for the maintenance of more consistent milk fat production. Dohme et al. (2008) found that cows fed less dietary peNDF had lower rumen pH than control cows. In another study, decreasing forage particle size caused an increase in time throughout the day when pH was below 5.8 and a decrease in minimal daily average pH (Krause et al., 2002).

The mechanism by which this occurs is also related to the role of the rumen mat in initiating rumination. Increases in dietary peNDF cause linear increases in total ruminating and chewing time (Yang and Beauchemin, 2006). Greater chewing activity increases the secretion of salivary $NaHCO_3$, which is attributed with buffering rumen digesta (Krause et al., 2002) and decreasing the risk for ruminal acidosis (Yang and Beauchemin, 2006). Proper balance of dietary peNDF may not always adequately maintain rumen pH; it is possible that low pH is caused by excessive dietary NFC or NSC instead of too little fiber (Mertens, 1997). Ruminal pH is affected by corn fermentability in addition to peNDF (Krause et al., 2002).

1.5. Feed Sorting in Dairy Cows

Cattle require sufficient levels of physically effective fiber in their diets for proper rumen function. If forage and diet particle size are too long, cows are able to sort out and refuse the large particles, and hence NDF and peNDF (DeVries et al., 2007), thereby not allowing them to

consume a balanced diet (Stone, 2004). This is especially problematic when feeding a low-forage diet (DeVries et al., 2009). Sorting activity has been observed throughout entire 24 h periods, including during the first 8 h after feeding (Kononoff et al., 2003b). It has been reported that cows are able to adjust their sorting behavior in only one day when presented with a dietary change (DeVries et al., 2007).

Competition at the feed bunk is greatest following the delivery of fresh feed. If fed a low forage diet, dominant cows may be sorting out and consuming large amounts of more rapidly fermentable carbohydrates (DeVries et al., 2007). Subordinate cows, who gain access to feed at a later time, may be forced to consume forage particles refused by the remainder of the herd. This scenario puts dominant cows at risk for SARA (DeVries et al., 2007) and subordinate cows at a potential risk for energy deficiency. Stone (2004), in his review, suggested that it would be unlikely for cows to preferentially consume long particles in an attempt to alleviate SARA.

Sorting behavior may be minimized by reducing the particle size of the diet. A reduction in corn silage particle size leads to greater consumption of coarse, high fiber particles and linearly decreases the NDF content of refusals (Kononoff et al., 2003b). Delivery of fresh feed to the bunk at least twice daily also reduces sorting, as well as improves access to feed and reduces variation in the composition of TMR consumed between cows (DeVries et al., 2007). It is possible that cows sort for a certain mean particle length, and that sorting in general may be able to be eliminated if a desirable geometric mean particle size is identified and used in TMRs (Maulfair et al., 2010).

1.6. Subacute Ruminal Acidosis

Inconsistent delivery of nutrients causes pH fluctuations and overall lower pH in the rumen (Dohme et al., 2008). Normal pH generally ranges from 6.6 before feeding to about 5.3 during periods of intensive fermentation in high producing cows fed high concentrate diets. The average daily rumen pH for high producing lactating dairy cows is often 6.0-6.2 (Zebeli et al.,

2010). SARA is a condition in which the pH of the rumen falls below 5.8 for an extended period of time (Stone, 2004; Yang and Beauchemin, 2006). This number has been chosen as the threshold for SARA because pH<5.8 is has been shown to be harmful to cellulytic bacteria (Russell and Wilson, 1996); acute acidosis begins at pH 5.2 (Stone, 2004). Age, genetics, inherent microbial population, and previous exposure to acidosis all affect the interaction between cows and the factors that lead to predisposition for SARA (Dohme et al., 2008).

The risk of SARA is increased by the accumulation of VFA from the fermentation of highly digestible carbohydrates. This is common when cows are moved from high- to low-forage diets at calving, and is enhanced by feed sorting. Consequently, cows in early lactation appear to be most susceptible to SARA (DeVries et al., 2009). High-producing cows typically have lower baseline pH, which contributes to the severity of the disease (Dohme et al., 2008). In addition, salivary $NaCO_3$ may not be able to buffer the large amounts of acid produced in the rumen of cows with high feed intake (Stone, 2004). These cows tend to experience longer and more severe bouts of SARA (Dohme et al., 2008).

Major strategies used to decrease the risk of SARA are increasing forage particle length and the amount of forage in the diet (Dohme et al., 2008). However, increasing the particle lengths of dietary forage does not increase chewing enough to increase salivary secretions significantly enough to affect ruminal pH (Kononoff et al., 2003b). Increases in peNDF cause an increase in time spent eating via eating rate and meal duration; rate of salivary secretion during eating is 1.3-2 times greater than resting salivation rate. An increase in daily time spent eating would increase saliva secretion and decrease the risk of SARA (Beauchemin et al., 2008).

Stone (2004) reported that VFA absorption, passage, and ruminal saliva dispersement are all increased with increasing rumen contractions, and that an increase in ruminal pH is associated with each of these processes. Heat stress can alter meal patterns to the extent that rumination and

rumen contractions are affected (Stone, 2004), increasing the risk for SARA. Decreasing eating rate in high-producing cows may also contribute to less risk of SARA by increasing the amount of saliva produced (Beauchemin et al., 2008).

The severity of SARA increases with each subsequent bout that the animal experiences (Dohme et al., 2008; DeVries et al., 2009). Minimum pH decreases, as well as the length of time spent at low pH (Dohme et al., 2008). Long term affects on cow behavior and rumen health have also been related to SARA. DeVries et al. (2009) observed an increase of greater than 2 h in feeding time from baseline on d 1 following the first bout of SARA, and only 1 h increases during subsequent rumen challenges. SARA can lead to rumenitis, and has the potential to create abnormalities in rumen papillae. This can decrease the absorptive capacity of the rumen wall, and consequently have long term negative effects on rumen health and milk production (Dohme et al., 2008). Krause and Oetzel (2005) suggested that a reduction in milk fat percentage probably does not occur following a single bout of SARA, but begins after multiple bouts. An acetate:propionate ratio < 2 is associated with SARA and may be responsible for decreased milk fat production (Krause and Oetzel, 2005).

1.7. Objective

The objective of this study was to gain a better understanding of feed sorting behavior in lactating dairy cows. It examined the effects of changes in rumen pH associated with SARA on individual-cow preferences for TMRs of varying corn silage particle size and carbohydrate fermentability.

2. Materials and Methods

2.1. Experimental Design and Sample Analysis

All animals used in this study were cared for according to guidelines established by The Pennsylvania State University Institutional Animal Care and Use Committee; all experimental

procedures involving the animals were approved. Eight Holstein cows weighing 682 ± 65 kg, averaging 219 ± 61 DIM, with parity 3.13 ± 0.99 (mean ± SD), and that had been previously ruminally cannulated were used in this study. Each cow was offered a long, slow fermenting and a short, fast fermenting ration ad libitum that were formulated to meet or exceed NRC (2001) recommendations. Diets contained corn grain, soybeans, canola meal, alfalfa and corn silage, and vitamins and minerals (Table 1), and were formulated to contain 16.5% CP, 29.5% NDF, and 45.7% NFC (Table 2). All animals were housed in individual stalls, and provided ad libitum access to water. Cows were milked twice daily at 0500 and 1700 h.

Animals were fed once daily at approximately 0730 h and allowed ad libitum access to each ration throughout the day. Dividers were placed between cows and in front of each cow to prevent the mixing of different TMRs. The side of delivery for each ration in the bunk for each cow was alternated daily. Cows were adapted to this feeding style for 7 d.

Differences in TMR particle size were created by utilizing corn silage of varying particle lengths in each ration. When corn was harvested for silage, several knives were removed from the self-propelled forage harvester (John Deere, model 6750) in order to produce extra long particles (theoretical cut length = 47 mm). Chopped corn was ensiled in an ag-bag (Ag-Bag, Miller-St. Nazianz, Inc., St. Nazianz, WI) for at least 4 months prior to feeding. Long silage was forced through a custom-made chopper 2 times to produce short silage on a daily basis, so that the fermentation profiles of different-sized silage would be equivalent. Differences in starch fermentability were created by using either coarsely (slow fermenting) or finely (fast fermenting) ground corn grain in each ration. Each ration was mixed separately using an I. H. Rissler model 1050 TMR Mixer (E. Rissler Mfg. LLC, New Enterprise, PA).

Forage (Table 3), ration, and refusal particle size distributions were determined by sieving samples in the American Society of Agriculture and Biological Engineers (ASABE) forage particle separator. Six fractions were determined for each sample: >26.9, >18.0, >8.98,

>5.61, >1.65, and <1.65 mm (ASABE., 2007). TMR particle size differed at each fraction except >8.98 mm (Table 4). The $peNDF_{<1.18}$ of the long and coarse (**LC**) ration was 25.2; short and fine (**SF**) TMR had 21.5 $peNDF_{<1.18}$. Dry matter percent of each sample was obtained following sieving by placing each sample in a forced air oven at 55° C for 48 h.

Feed intake was monitored beginning on d 8. Intakes from d 8, 9, 10, and 11 were averaged to establish baseline intake for each ration. On d 11, rumen contents were collected from dorsal, ventral, cranial, caudal, and medial areas of the rumen at 0.0, 1.5, 3.5, 5.5, 8.5, 11.5, 14.5, 18, 21.5, and 24 h after feeding (Kononoff et al., 2003b) to determine baseline rumen conditions. Digesta were mixed by hand and filtered through 4 layers of cheesecloth. pH of the rumen fluid was measured immediately using a handheld pH meter (HI 98121, HANNA Instruments Inc., Woonsocket, RI). Approximately 15 ml of fluid was placed into bottles containing 3 ml of 25% metaphosphoric acid and 3 ml of 0.6% 2-ethylbutyric acid (internal standard) and stored at -20° C. Samples were later thawed and centrifuged for 30 minutes at 4000 x g at 4° C 3 times and supernatants were analyzed for VFA concentration using gas chromatography (Yang and Varga, 1989).

On d 12, feed was restricted to 75% of baseline intake. One h before feeding on d 13 (challenge d), approximately 4 kg (as-fed) of finely ground wheat grain was mixed thoroughly into the rumens of all cows. Each cow was then allowed ad libitum access to each TMR for the remainder of the trial. Rumen contents were sampled as described above at 11.5, 14.5, 18, and 21.5 h post-feeding on feed restriction d; 0.0, 1.5, 3.5, 5.5, 8.5, 11.5, 14.5, 18, 21.5, and 24 h after feeding on challenge d; and 3.5, 8.5, 14.5, and 21.5 h after feeding on recovery d.

2.2. Statistical Analyses

Statistical analysis was conducted using PROC MIXED of SAS (2006). Dependent variables were analyzed as a cross over design. All denominator degrees of freedom for *F*-tests were calculated according to Kenward and Roger (1997) and repeated measurements for rumen

pH and VFA concentrations were analyzed using the first order autoregressive covariance structure (Littell et al., 1998), as well as terms for time and interaction of treatment by time. Because of unequally spaced rumen sampling, the weighted mean daily pH and VFA concentrations were determined by calculating the area under the response curve according to the trapezoidal rule (Shipley and Clark, 1972). For each cow, the 4 control d (8, 9, 10, and 11) were averaged before analysis to provide equal number of observations between control and rumen challenge d. Samples were analyzed for effects of day, diet, and their interaction. A selection index based on refusals was calculated for each of the 6 particle size fractions. This index was calculated as the actual intake of each fraction (Y_i to pan) expressed as a percentage of the expected intake. Expected intake of Y_i equals intake multiplied by the fraction of Y_i in the fed TMR (Leonardi and Armentano, 2003). Values > 1 indicate cows were sorting for the particle fraction and values < 1 indicate cows were sorting against the particle fraction. The 95% confidence limits were used to determine if selection index was significantly different than 1. All data are presented as least squares means and treatment effects were considered significant when $P < 0.05$ and a trend when $P < 0.10$.

3. Results and Discussion

Cows were given ad libitum access to both TMRs following the administration of wheat grain. To ensure that the amount of each TMR in their bunk did not provide any limitations on intake, 110% of total DMI of each ration was delivered to each cow on challenge d. Because of this, there were large differences in refusal weights collected on the final baseline d versus any d in the challenge procedure (feed restriction, P = 0.002; challenge, < 0.0001; recovery d 1, 0.0028; recovery d 2, 0.0051). Individual cow decisions to consume either ration as-desired were unaffected by the amount of either TMR in the bunk.

There was no difference in total DMI between days (P = 0.1074). This was also shown by Dohme et al. (2008). There was an effect of diet chosen and consumed by the cows (P < 0.0001), with large differences between days of the study (P = 0.0082). There was no effect of side of feed delivery (P = 0.6832) in the bunk or side of the automatic water bowl in the stalls (P = 0.6259) on intake or preference for either ration**.**

There was a two-fold increase in the percent of total DMI consumed as LC TMR on challenge d. Baseline preference for LC TMR was 18.1%; this increased to 38.3% on challenge d (P < 0.0001; Table 5). All preferences were returned to baseline by d 2 following the challenge (LC, P = 0.9229; SF, P = 0.4481). Preferences for each ration for the duration of the challenge procedure are presented in Figure 1.

Allowing each cow access to 2 separate TMRs with significantly different particle size distributions (Table 4) attempted to eliminate feed sorting so that the effect of SARA on preferred diet composition could be determined. Minimal sorting of each ration was observed, however (Table 6). Sorting for different particle fractions occurred to the same extent in both rations for all fractions except particles < 1.65 mm. This can be explained by slight, insignificant differences in sorting at all other particle fractions, resulting in an increase in accumulated particles in the pan for SF refusals. It is not believed that the low-level sorting activity exhibited in this study played any role in or represented the cow response to the rumen challenge.

The weighted mean baseline rumen pH was 6.02, which is consistent with the average pH value as reviewed by Zebeli et al. (2010). The weighted mean pH on challenge d was 5.77, a significant decrease from baseline (P < 0.0001). Ruminal pH patterns varied throughout the d for baseline and challenge d (Figure 2). DeVries et al. (2009) considered pH 5.8 total SARA and 5.5 severe SARA. By this definition, cows in the present study were not affected by severe acidosis. The increase in consumption of LC TMR was associated with the decrease in mean rumen pH. This would suggest that cows may selectively consume more $peNDF_{<1.18}$ when experiencing a

bout of SARA. Krause et al. (2002) reported that a decrease in forage particle size led to a decrease in rumen pH. This may explain why the consumption of the SF ration decreased during the challenge.

DeVries et al. (2008) observed feed sorting to a greater extent in cows considered to be at high risk for SARA than in those considered to be at low risk. This suggests that cows experiencing lower ruminal pH may choose to consume increased amounts of TMR of sufficient $peNDF_{<1.18}$ as necessary to re-establish a proper rumen environment. This idea is supported by the large increase in preference for LC TMR in the present study. The present data is also in agreement with findings by Keunen et al. (2002).

The change in microbial fermentation patterns induced by feed restriction followed by the introduction of highly fermentable ground wheat grain into the rumen of each cow caused a change in the normal fluctuation of VFA concentrations. Weighted averages were calculated for the concentration of primary VFAs for baseline and challenge d (Figure 3); ranges are represented by minimum and maximum values (Table 7).

A difference was found in acetic ($P = 0.0083$) and butyric ($P = 0.0027$) acid concentrations between baseline and challenge d. There was no difference in mean propionic acid concentration between d ($P = 0.3886$), but variation in levels was observed at different times postfeeding (Figure 3). Ranges were significantly different between d for acetic and butyric acids (Table 7). The most relevant effect of SARA on VFA concentration is the change in range and concentration of acetic acid, which serves as an important substrate for mammary fatty acid synthesis (Annison et al., 1968)

Mertens (1997) wrote that decreases in A:P may be linked to reduced milk fat production, which has a direct economic impact in the dairy industry. A:P on challenge d was significantly higher than baseline ($P = 0.0303$) in the present study (Figure 4). The weighted mean ratio changed from 2.3 to 2.6, and the minimum daily ratios were 1.90 and 2.03 ($P = 0.2330$). Krause

and Oetzel (2005) used 2.0 as the threshold for decreases in milk fat production. According to this, the risk for negative effects on milk components was not altered by the rumen challenge.

4. Conclusion

The objective of this study was to determine whether cows changed their preference for 2 different TMRs of varying particle size and fermentability while experiencing SARA. Increases in preference for TMR of long particle size and slow fermenting starch ($P < 0.0001$) accompanied the decreases in rumen pH ($P < 0.0001$) and changes in VFA concentration (acetic, $P = 0.0083$; butyric, $P = 0.0027$) associated with SARA. It is inferred from these findings that cows can selectively increase their intake of $pdNDF_{<1.18}$ during a bout of SARA.

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Appendix I

Table 1. Physical composition of formulated rations.[1]

Item	% DM
Corn Grain[2]	22.16
Roasted Soybeans[3]	7.07
Canola Meal	9.38
Alfalfa Haylage	15.39
Corn Silage[4]	42.63
Vitamin and Mineral Mix	2.52
NaCl	0.43
Optigen®[5]	0.41

[1]Rations varied in processing of corn grain and particle size of corn silage.
[2]Coarse-ground corn grain was used in the long and coarse ration; fine-ground corn was used in the short and fine ration.
[3]Roasted soybeans were split; hulls were removed.
[4]Long corn silage was produced by removing knives from the chopper at harvest and included in the long and coarse ration; long silage was forced through a custom-made chopper twice to produce short corn silage with an equivalent fermentation profile for the short and fine ration. Particle size distributions are shown in Table 3.
[5]Optigen® = Controlled release non-protein nitrogen.

Table 2. Chemical composition of ration[1] as-formulated using CPM-Dairy.

Nutrient	Composition, % of DM
CP	16.50
ADF	19.16
NDF	29.53
Forage NDF	23.24
Ether Extract	5.24
Ash	5.85
NFC[2]	45.72
NE_L, Mcal/lb	1.69

[1]Rations varied in processing of corn grain and particle size of corn silage. Chemical profile of both rations were equivalent.
[2]NFC=nonfiber carbohydrate. Calculated by difference: 100 – (% CP + % NDF + % Fat + % Ash).

Table 3. Particle size distributions of forages determined with ASABE particle separator for TMR.

Item	Long Corn Silage	Short Corn Silage	Alfalfa Haylage
Particle size, as-fed % retained[1]			
26.9 mm	10.19	0.83	1.57
18.0 mm	32.61	12.45	7.15
8.98 mm	29.47	28.60	30.88
5.61 mm	14.22	19.91	24.50
1.65 mm	12.59	30.50	30.33
Pan	0.92	7.71	5.57
DM, %	38.63	40.92	39.46

[1]Approximate equivalency to Penn State Particle Separator (PSPS): top sieve (26.9 + 18.0 mm), middle sieve (8.98 mm), lower sieve (5.61 + 1.65 mm), and pan (pan) (Maulfair et al., 2010).

Table 4. Particle size distributions determined with ASABE particle separator for TMR containing long corn silage particles and coarse ground corn or short corn silage particles and fine ground corn.

Item	Long and Coarse	Short and Fine	SEM	P-value
Particle size, as-fed % retained[1]				
26.9 mm	3.20	0.65	0.15	<0.0001
18.0 mm	15.33	4.19	0.27	<0.0001
8.98 mm	20.23	17.12	0.76	0.0092
5.61 mm	22.54	17.47	0.65	<0.0001
1.65 mm	24.00	33.45	0.52	<0.0001
Pan	14.69	27.11	1.18	<0.0001
$peNDF_{1.18}$[2]	25.19	21.52	0.35	<0.0001
DM, %	49.65	50.07	0.46	0.5232

[1]Approximate equivalency to Penn State Particle Separator (PSPS): top sieve (26.9 + 18.0 mm), middle sieve (8.98 mm), lower sieve (5.61 + 1.65 mm), and pan (pan) (Maulfair et al., 2010).
[2]Physically effective $NDF_{1.18}$ = % of particles >1.65 mm x NDF of whole sample (similar to top 3 sieves in PSPS) (Kononoff et al., 2003a).

Table 5. Intake[1] of TMRs[2] of different particle size and fermentability during a rumen challenge.

Item	DMI	SEM	P-value
Long[3] and Coarse[4]			
Baseline	5.30[a]	1.77	0.0038
Feed Restriction d	4.04[a]	1.77	0.0256
Challenge d	12.57[b]	1.77	<0.0001
Recovery d 1	7.81[a]	1.77	<0.0001
Recovery d 2	5.06[a]	1.77	0.0056
Short[3] and Fine[4]			
Baseline	25.43[a]	1.77	<0.0001
Feed Restriction d	19.32[b]	1.77	<0.0001
Challenge d	20.22[b]	1.77	<0.0001
Recovery d 1	21.64[a]	1.77	<0.0001
Recovery d 2	23.52[a]	1.77	<0.0001

[a-b]Means within each ration with different superscripts differ (P < 0.0001).
[1]As-fed.
[2]Rations varied in particle size of corn silage and processing of corn grain.
[3]Long corn silage was produced by removing knives from the chopper at harvest and included in the long and coarse ration; long silage was forced through a custom-made chopper twice to produce short corn silage with an equivalent fermentation profile for the short and fine ration. Particle size distributions are shown in Table 3.
[4]Coarse-ground corn grain was used in the long and coarse ration; fine-ground corn was used in the short and fine ration.

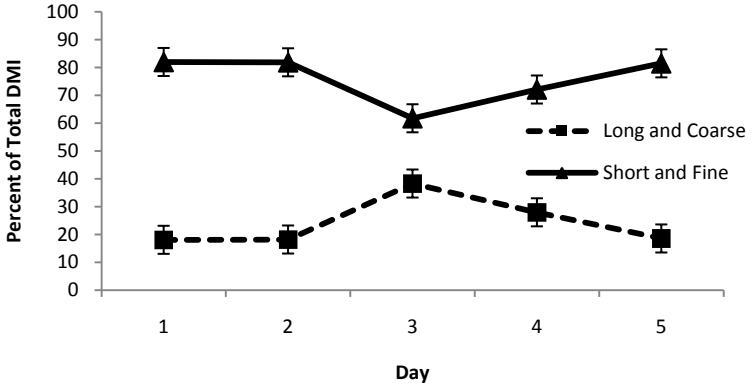

Figure 1. Percent of total DMI consumed as long and coarse or short and fine TMR during a rumen challenge. Baseline = d 1, feed restriction = d 2, challenge = d 3, recovery = d 4 and 5.

Table 6. Sorting indices[1] for TMRs[2,3] of different particle size and fermentability

Item	Long and Coarse	Short and Fine	SEM	P-value
Particle size, as-fed % retained[4]				
26.9 mm	0.78	0.98[5]	0.05	0.0072
18.0 mm	0.85	0.90	0.03	0.2053
8.98 mm	0.91	0.90	0.01	0.0056
5.61 mm	1.03	1.00[5]	0.01	0.0192
1.65 mm	1.06	1.01[5]	0.01	0.0031
Pan	1.15	1.02[5]	0.02	< 0.0001

[1]1.0 indicates no sorting; values < 1.0 indicate sorting against, values .> 1.0 indicate sorting in favor.
[2]Rations varied in particle size of corn silage and processing of corn grain (as-delivered).
[3]Particle size distribution of refusal samples were determined with ASABE particle separator for TMR.
[4]Approximate equivalency to Penn State Particle Separator (PSPS): top sieve (26.9 + 18.0 mm), middle sieve (8.98 mm), lower sieve (5.61 + 1.65 mm), and pan (pan) (Maulfair et al., 2010).
[5]Value is statistically equivalent to 1.0 based on a 95% confidence interval.

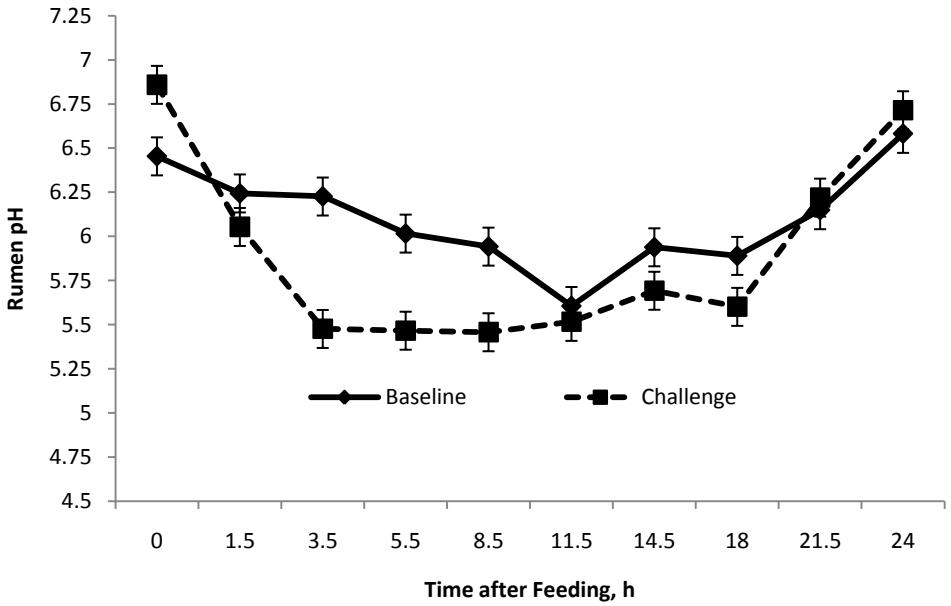

Figure 2. Ruminal pH of lactating dairy cows undergoing a rumen challenge. Digesta were filtered through cheesecloth; pH of filtered liquid was measured.

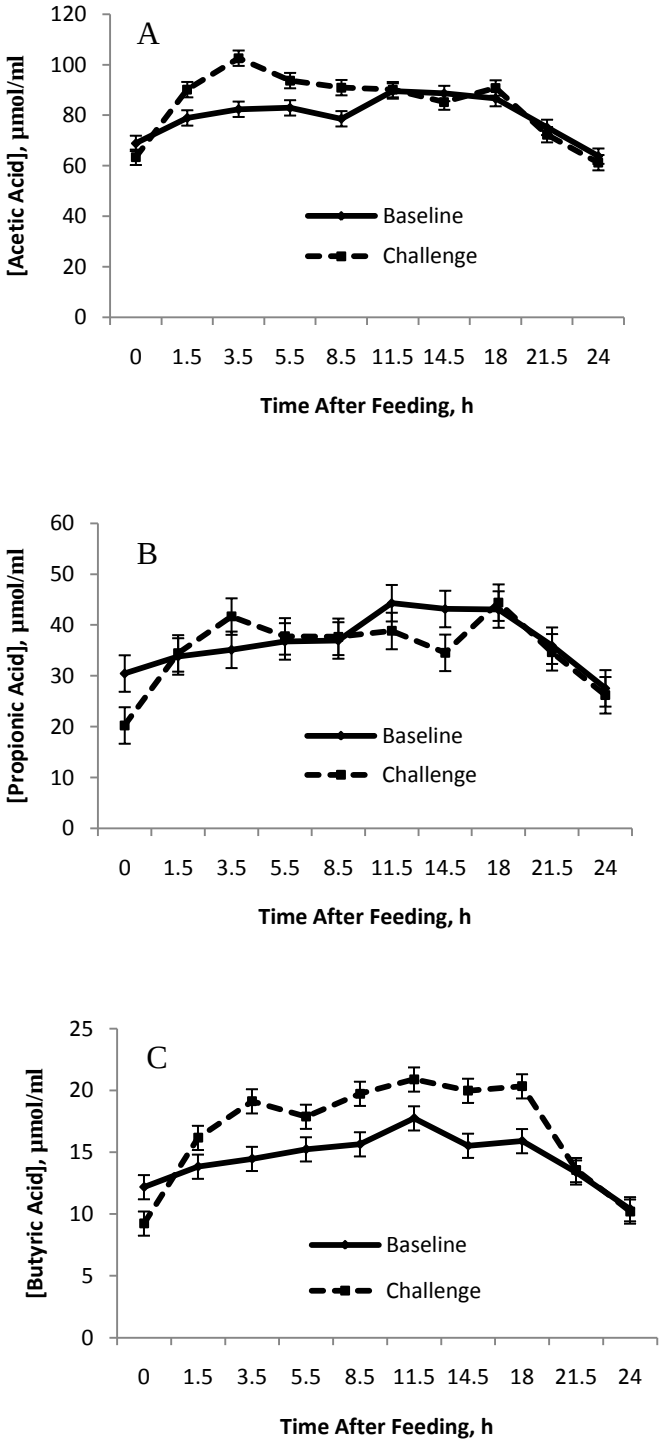

Figure 3. Concentration of acetic (A), propionic (B), and butyric (C) acids in the rumens of lactating dairy cows undergoing a rumen challenge. Concentrations were determined using gas chromatography (Yang and Varga, 1989).

Table 7. Differences in range of primary VFA concentrations in the rumens of lactating dairy cows during a rumen challenge.[1]

Item	Acetic		Propionic		Butyric	
	Min.	Max.	Min.	Max.	Min.	Max
Baseline	62.24	96.16	26.73	48.65	10.37	18.62
Challenge	58.31	106.94	19.97	47.42	8.96	24.14
SEM	2.61	2.03	1.91	3.98	0.59	0.99
P-value	0.0083	0.0021	0.0006	0.6763	0.0077	0.0018

[1]Samples were collected at 0.0, 1.5, 3.5, 5.5, 8.5, 11.5, 14.5, 18, 21.5, and 24 h after feeding.

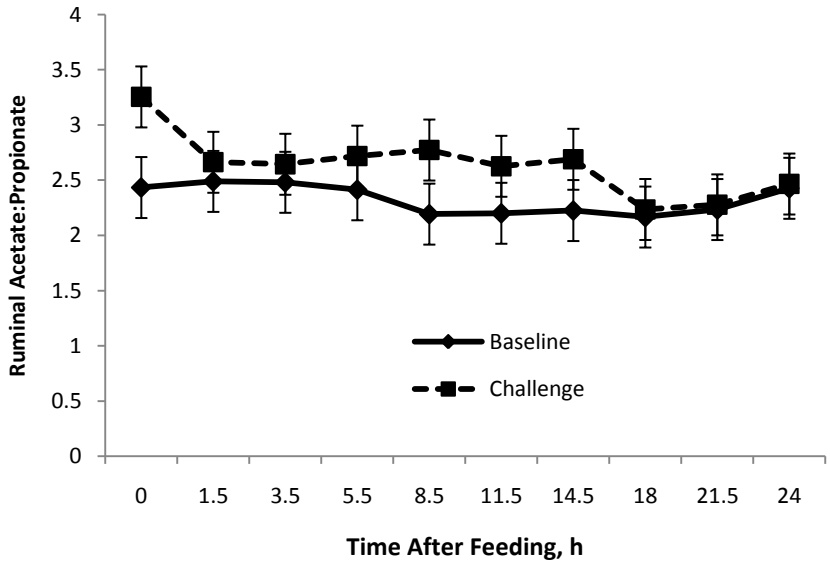

Figure 4. Ratio of acetic and propionic acids in the rumens of lactating dairy cows during a rumen challenge. A difference existed between weighted average A:P on baseline and challenge d (P = 0.0303).

Appendix II

Cows were housed in a tie stall barn, with automatic water bowls and ad libitum access to feed. Dividers were placed in the feed bunk between cows and in front of each cow to prevent the mixing of TMRs.

Cows were offered long and slow fermenting (**Left**) and short and fast fermenting (**Right**) TMR.

Custom-made forage chopper and Rissler TMR mixer. Long corn silage was forced through this chopper twice to produce short silage. This was done each time feed was mixed.

Kolby McIntyre collecting rumen digesta samples through a rumen cannula.

A few of the people who worked on this trial. Left to right: Dr. Laming Ding, Daryl Maulfair, Kolby McIntyre, Hilary Pasi, and Catherine Claxton. See 'Acknowledgements' for a complete list of people involved with the study.

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EXPERIENCE

Penn State University Beef Cattle and Sheep Center University Park, PA
Student Worker *01/2010 – Present*

- Feed Black Angus cattle and Dorset sheep on a regular basis.
- Install controlled internal drug release devices (CIDRs) in cattle; observe for heat.
- Administer vaccines and estrous synchronization injections to cattle
- Assist ewes in distress while lambing; bottle feed triplet lambs.

Russell Veterinary Hospital Russell, PA
Veterinary Assistant (Part Time) *05/2008 – Present*

- Draw blood; perform laboratory analysis upon blood and urine.
- Prepare animals for surgical procedures; assist the surgeon and anesthesiologist.
- Responsible for the organization of radiographs taken by hospital staff.

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American Society of Animal Science Undergraduate Scholarship Recognition Award, April 2010
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