Effects of ad libitum or restricted access to total mixed ration with supplemental long hay on production, intake, and rumination

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ABSTRACT

Limiting feeding time has been a concept used in growing and nonlactating ruminant animals with good success, especially in improving feed efficiency while maintaining normal rumen function and fiber digestibility. This study evaluated the physiological and productive responses of cows fed a total mixed ration (TMR) available for 24 or 19 h/d with or without access to additional long hay. Eight multiparous Holstein cows were used in a replicated 4 × 4 Latin square design with 21-d periods. Rations were formulated to mimic a TMR used in the Parmigiano Reggiano cheese production area of Italy, consisting of all dry and nonfermented components. Intakes were reduced by 2.49 kg/d in cows with restricted TMR access and 1.16 kg/d without supplemental hay. Rumen characteristics were similar for all groups with improvements in energy-corrected milk. The results show that once cows adapted to diet changes, few differences were observed when offering TMR continuously or with limited access. Energy-corrected milk feed efficiency was improved in cows having access to feed 19 versus 24 h/d. In addition, the presence of long hay during the TMR restriction offered the optimal conditions for ruminal function and pH stability, yet no effects were observed on body weight change. A Latin square with 21-d periods could have a carry-over effect on energy storage, and mobilization of fat reserves might be able to mask negative energy balance during restriction. Restricted feeding could be used as a strategy to manage feed availability according to cow production and metabolic condition, to maximize the use of nutrient resources, reducing the cost of milk production and improving the cows' welfare and health.

Key words: limit feeding, lactating dairy cow, rumination

INTRODUCTION

Total mixed rations with dry hay as the only forage source are widely used in the Parmigiano Reggiano production area of Italy. Typically, little to no water is added to the TMR to avoid the risk of fermentation in the manger. In this situation, particle size of the feed must be reduced to prevent feed sorting, and therefore physically effective NDF (**peNDF**) is generally below what is commonly considered acceptable (Fustini et al., 2016). This may lead to a reduction of rumination time and saliva secretion thereby leading to SARA (Khafipour et al., 2009). Italian hays are often of low quality and reduced nutrient value due to the climatic conditions (Palmonari et al., 2016), which often necessitates an increased amount of concentrate in the diet to satisfy the energy requirements of lactating cows. Adequate forage particle size in the ration is therefore critical to maintain rumen function and milk components. Furthermore, peNDF promotes chewing and salivary secretion, and maintaining rumen conditions for VFA absorption, which improves pH stability in the rumen (Mertens, 1977, 1997). In a recent study, Kmicikewycz and Heinrichs (2014) concluded that supplementing rations with long hay favored the recovery of animals with SARA in early and mid lactation. Moreover, inclusion of long hay in low proportions, particularly in diets based on finely chopped silages, has been shown to improve rumination time (Beauchemin et al., 1994).

Dairy cows may also be able to select in favor of longer particles if rumen pH and rumination levels are low. Maulfair et al. (2013) saw that during SARA cows were able to change their eating preferences in favor of diets with more long forage and lower contents of fermentable starch. Keunen et al. (2002) also showed that lactating cows with induced SARA increased their preference for long alfalfa hay compared with alfalfa pellets. It has been suggested that under conditions of reduced ruminal pH, if given an option, cows will choose feeds with more buffering capacity (DeVries et al., 2008).

Limit feeding has been used in ruminant animals with good success, especially in improving feed effi-

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ciency while maintaining normal rumen function and digestibility (Owens et al., 1995). Limit feeding often improves digestibility (Zanton and Heinrichs, 2008), primarily through reducing the rate of passage of material from the reticulo-rumen of animals that are fed less (Bhatti et al., 2008). However, few trials based on this concept have been conducted with cows in mid lactation.

The purpose of this experiment was to evaluate the physiological and productive response in cows fed a dry, finely chopped TMR administered for a 24 or 19 h/d, with or without additional free choice long hay. The hypothesis was that cows having limited access to TMR would result in faster eating rates and lead to changes in rumen pH and that offering long hay in addition would stabilize rumen pH and production.

MATERIALS AND METHODS

This study was conducted at the University of Bologna (Italy), and all experimental procedures involving animals were approved by the University of Bologna Animal Care and Use Committee.

Experimental Design and Data Collection

Eight multiparous Holstein cows (average BW $\pm$ SD: 629 $\pm$ 49 kg) were blocked by parity (2.25 $\pm$ 0.46), milk production (44.9 $\pm$ 2.5 kg/d), and DIM (99 $\pm$ 49) and used in a replicated 4 $\times$ 4 Latin square design study with 21-d periods (14 d adaptation and 7 d collection). Rations were formulated using DinaMilk5 (Faberma, Ostiano, Italy) to mimic TMR used in the Parmigiano Reggiano cheese production area of Italy and consisted of all dry and nonfermented components. The basal diet was the same for all 4 treatments with or without access to additional long grass hay, which was the same source of hay used in the TMR (Table 1). Diets were offered in individual feed mangers divided in 2 equal parts, one for the TMR and the other for the long grass hay. Cows were offered ad libitum intake of TMR for 24 or 19 h/d; TMR (approximately 1.10 $\times$ expected intake) both were fed once a day at 2000 h (Zago Mixer, Padova, IT). The study was conducted during the months of September through December, and feeding was done at a time to encourage intake in the evening hours. Animals were housed in a naturally ventilated tiestall barn and milked twice a day (0800 and 1930 h) in a double-5 herringbone milking parlor. Treatments were identified by the length of TMR access as 24 or 19 h, with (F+) or without (F−) ad libitum long grass hay.

From d 15 to 21 of each period, we collected data and took samples. Body weight was measured daily

(Afiweight scale, Afikim, Israel), and DMI was determined by recording feed offered and refused for each cow. Samples of diets and orts were collected daily and a portion of each sample was dried in a forced-air oven at 105°C for 24 h for DM determination. Milk yield was recorded daily (Afimilk Information Management System, Afikim, Israel) during d 15 to 21 of each period. Milk samples from 2 consecutive milkings for each cow were collected on d 17 and 18 of each period, preserved (Bronolab-W II Liquid Preservative; D & F Control Systems Inc., Dublin, CA), and analyzed for fat, true protein, and lactose by infrared spectroscopy

Table 1. Composition of the TMR diet and the hay (mean $\pm$ SD)

Item	TMR ¹	Hay
DM, %	87.66 $\pm$ 0.72	88.54 $\pm$ 2.93
Ether extract, % of DM	2.53 $\pm$ 0.38	1.64 $\pm$ 0.20
Ash, % of DM	9.76 $\pm$ 0.37	8.93 $\pm$ 0.63
aNDFom, ² % of DM	31.44 $\pm$ 5.45	58.46 $\pm$ 3.45
ADF, % of DM	20.77 $\pm$ 4.18	42.61 $\pm$ 1.62
ADL, % of DM	2.94 $\pm$ 0.64	6.62 $\pm$ 0.80
IVNDFD ³ 24 h, % of aNDFom	77.33 $\pm$ 6.95	67.02 $\pm$ 3.21
d-NDF 24 h, ⁴ % of aNDFom	45.51 $\pm$ 3.44	44.74 $\pm$ 5.27
uNDF ₂₄₀ , ⁵ % of DM	6.40 $\pm$ 1.98	17.82 $\pm$ 5.40
Starch, % of DM	23.55 $\pm$ 4.78	1.98 $\pm$ 2.12
Sugar, % of DM	6.96 $\pm$ 0.56	4.79 $\pm$ 0.66
CP, % of DM	14.28 $\pm$ 0.90	8.76 $\pm$ 1.10
Soluble protein, % of DM	3.91 $\pm$ 0.44	3.55 $\pm$ 0.37
NDIP, ⁶ % of DM	3.00 $\pm$ 0.28	4.92 $\pm$ 0.65
ADIP, ⁷ % of DM	1.01 $\pm$ 0.07	1.55 $\pm$ 0.15
PSPS ⁸		
19 mm, %	0.39 $\pm$ 0.79	
8 mm, %	15.18 $\pm$ 5.10	
4 mm, %	18.73 $\pm$ 1.92	
Bottom, %	65.69 $\pm$ 5.60	
Ro-Tap, % >1.18 mm	46.24 $\pm$ 3.90	
peNDF, ⁹ % of DM	14.14 $\pm$ 1.30	

¹TMR ingredients: 34.5% grass hay, 19.2% steam flaked corn, 3.9% cane-beet molasses blend, and 42.4% grain mix {29.6% wheat bran, 29.4% sorghum grain, 21.6% soybean meal, 14.7% flaked soybean, 2.2% calcium carbonate, 1% sodium chloride, 0.4% magnesium oxide, 0.9% sodium bentonite, and 0.3% vitamin and mineral premix [provided 40,000 IU of vitamin A, 4,000 IU of vitamin D₃, 30 mg of vitamin E 92% α -tocopherol, 5 mg of vitamin B₁, 3 mg of vitamin B₂, 1.5 mg of vitamin B₆, 0.06 mg of vitamin B₁₂, 5 mg of vitamin K, 5 mg of vitamin H₁ (para-aminobenzoic acid), 150 mg of vitamin PP (niacin), 50 mg of choline chloride, 100 mg of Fe, 1 mg of Co, 5 mg of I, 120 mg of Mn, 10 mg of Cu, and 130 mg of Zn]}.

²Amylase- and sodium sulfite-treated NDF with ash correction.

³In vitro NDF digestibility, in % aNDFom.

⁴Digestible NDF at 24 h.

⁵Unavailable NDF estimated via 240-h in vitro fermentation.

⁶Neutral detergent insoluble protein (neutral detergent insoluble nitrogen $\times$ 6.25).

⁷Acid detergent insoluble protein (acid detergent insoluble nitrogen $\times$ 6.25).

⁸Penn State Particle Separator, % sample retained on each sieve (Lammers et al., 1996).

⁹Physically effective NDF (aNDFom $\times$ physical effectiveness factor), calculated using the Ro-Tap (W.S. Tyler, Mentor, OH) system (Mertens, 1997).

procedures (Associazione Provinciale Allevatori Bologna, Foss 4000, Foss Technology). Feed efficiency (kg/kg) was calculated as milk/DMI, 3.5% FCM/DMI, and solids-corrected milk/DMI during the experimental week.

Rumen Characteristics

Cows were monitored for rumination activity from d 15 to 20 of each period using the Hi-Tag rumination monitoring system (SCR Engineers Ltd., Netanya, Israel). Data Flow software analyzed rumination time with a resolution of 2 h (Schirmann et al., 2009) and calculated the rumination time during the last 24 h. To continuously monitor rumen pH, all cows received an indwelling wireless pH-transmitting unit (SmaXtec Animal Care Sales GmbH, Graz, Austria), which has been validated with rumen-cannulated dairy cows (Klevenhusen et al., 2014). These units (3.5 cm i.d., 12 cm long, and weighing 210 g) were manually inserted into the reticulorumen via the esophagus on d 14 of the first period. Prior to this, the units were calibrated following the company's instruction protocol (www.smaxtec.com). The units measured pH and temperature every 10 min and transmitted the data in real time to a base station using the ISM band (433 MHz). Both data of pH and temperature were collected using an analog-to-digital converter and stored in an external memory chip. Data of pH from d 15 to 21 of each period were analyzed as daily mean pH and time (min/d) below specific cut-off points (5.5 and 5.8). Wireless sensors were located in the ventral reticulorumen (Gasteiner et al., 2009), and few differences were observed between pH measured by wireless units in the rumen and in the ventral rumen sac (Klevenhusen et al., 2014), the term "reticuloruminal pH" will be used.

On d 5 and 6 of experimental week, rumen fluid was collected via esophageal probe at 12 h (0800 h) and 24 h (2000 h) after feeding. The first 500 mL of rumen fluid collected was discarded before taking samples. Volatile fatty acid concentrations were determined by GC (Goetsch and Galyeen, 1983), whereas ammonia was evaluated via commercial kit (urea/BUN – color, BioSystems S.A. Barcelona, Spain) according to the producer procedure.

Chemical Analysis

Individual feed ingredients were collected weekly and dried in a forced-air oven at 55°C for 48 h for DM. During the experimental week of each period, diets were sampled daily and analyzed for chemical composition according to the following methods: CP; amylase-treated, ash-corrected NDF (aNDFom); and ADF according to

Mertens et al. (2002) and AOAC method 973.18 (AOAC International, 2016). Starch was determined according to AOAC method 996.11 and ether extract according to AOAC method 920.390020 (AOAC International, 2016). Forage and diet composite samples were used to determine particle size distribution on an as-fed basis using the Penn State Particle Separator (Lammers et al., 1996), physical effectiveness factor using a RoTap Separator (W.S. Tyler, Mentor, OH). Diet peNDF was calculated as the product of the total diet NDF content and its physical effectiveness factor (Mertens, 1997). In vitro digestibility (24, 48, and 240 h) of aNDFom of forage composite and diet composite samples (1-mm grind; Wiley mill; Arthur H. Thomas, Philadelphia, PA) were determined using an in vitro fermentation (Tilley and Terry, 1963) in buffered medium containing ruminal fluid (Goering and Van Soest, 1970). Ingredients and diets were also analyzed for in vitro aNDFom digestibility at 24 and 240 h according to the procedure described by Palmonari et al. (2017). Briefly, in vitro aNDFom digestibility at 24 and 240 h was performed using the Tilley and Terry modified technique (Tilley and Terry, 1963; Van Soest et al., 1991).

Statistical Analysis

Data for DMI, milk yield and composition, feed efficiency, microbial protein supply, and BW, and were analyzed as a replicated Latin square design with model effects for diet, period, and replicate using the MIXED procedure of SAS (version 9.1, SAS Institute Inc., Cary, NC) using cow within replicate as a random effect. Repeated measurements on performance data (i.e., DMI, milk yield, and so on) were reduced to period means for each cow before statistical analysis. Data for ruminal pH, NH₃N, and VFA were analyzed with repeated measures using the MIXED procedure of SAS. The model included effects of diet, period, time, and the interaction of diet and time, with cow as a random effect. Interaction terms were tested and found nonsignificant and therefore were not included in the final models except for rumination time, which is noted in the table. Least squares means were separated using PDIFF option with Tukey's adjusted *P*-values when a significant *F*-test ($P \leq 0.05$) was detected.

RESULTS AND DISCUSSION

The TMR fed was adequate in terms of nutrient composition (NRC, 2001) but smaller in particle size (peNDF = 14.14 ± 1.30%) than what is often recommended (peNDF of 21: Heinrichs, 2013; Kmicikewycz et al., 2015; Table 1). This level of dietary particle size minimized the risk of cows sorting (Maulfair et al., 2011;

Table 2. Effect of feed restriction and supplemental hay¹ on DMI and fiber fraction intake

Item	Feed availability				SEM	<i>P</i> -value	
	24 h		19 h			24 vs. 19 h	F+ vs. F–
	F+	F–	F+	F–			
TMR DMI, kg	27.36	24.60	23.27	23.71	1.66	0.04	0.31
Hay DMI, kg	0.55	—	0.70	—	0.09	0.09	—
DMI, kg	27.85	24.56	24.05	23.65	1.70	0.06	0.13
BW, kg	637.93	637.86	646.56	631.67	20.99	0.76	0.08
aNDFom, ² kg/d	8.79	7.66	7.73	7.44	0.54	0.10	0.07
aNDFom, % of BW	9.39	9.48	9.31	9.51	0.34	0.83	0.24
uNDF, ³ kg/d	1.89	1.60	1.70	1.57	0.12	0.17	0.02
uNDF, % of BW	0.30	0.26	0.26	0.25	0.02	0.25	0.19

¹F+ = supplemental hay; F– = no supplemental hay.

²Amylase- and sodium sulfite-treated NDF with ash correction.

³Unavailable NDF estimated via 240-h in vitro fermentation.

Fustini et al., 2016). When cows were offered TMR 24 vs. 19 h/d, TMR DMI increased (25.98 vs. 23.49 kg, SEM = 1.66, $P = 0.04$, Table 2). Supplemental hay intake was similar between diets. Consequently, total DMI tended to be greater for cows with 24-h TMR access (26.21 vs. 23.85 kg, SEM = 1.70, $P = 0.06$, Table 2) compared with cows offered TMR 19 h/d. Diets including hay tended to increase total NDF intake (8.26 vs. 7.55 kg/d, SEM = 0.54, $P = 0.07$, Table 2). When hay was offered in addition to TMR, cows consumed relatively little hay (<1 kg/d) and tended to consume more hay when TMR was offered for 19 versus 24 h/d. However, this hay intake was not equal to the reduction in TMR intake, likely due to feed palatability or preference for the TMR over the hay alone. A change in eating behavior may have occurred and may fall into the category of slug feeding once cows had adapted to this feeding (19 vs. 24 h) change (Fustini et al., 2016). According to other authors (Albright, 1993; Maulfair et al., 2013), animals modified their eating behavior after adaptation to a different feeding regimen. It was thought that when dairy cows had a limited amount of TMR, it would result in faster eating rates, which could lead to substantial diurnal changes in VFA production and increase the risk of SARA (Van Soest, 1994). Previous work (Kmicikewycz and Heinrichs, 2014) demonstrated that long hay availability with no TMR offered could decrease risk of SARA in lactating dairy cows. This hay availability should also limit the chance of a higher amount of feed being consumed in the first meal after TMR delivery the following day. This condition is often called “slug feeding,” and is characterized by a long period of time in which cows remain out of feed, after which they rapidly consume an amount of fermentable carbohydrates able to induce a drop in ruminal pH (Krause and Oetzel, 2005). Cows with TMR available for 19 h/d consumed 37.1% of their hay intake

in the 5 h after TMR was removed, whereas cows with TMR continuously available had 14.4% of their hay intake in the same period of time. Due to the quality of this hay, cows that had it available had higher intakes of NDF and uNDF. Results of fiber digestibility (total-tract digestibility >75% in all the treatments; Table 3) appeared to be comparable to values obtained in other experiments, and confirm digestibility shown by Fustini et al. (2017), given the quality of forage used in the dry diets of the Parmigiano Reggiano cheese production area of Italy (Palmonari et al., 2016).

No differences were observed in milk production between the 24- and 19-h feed groups; however, cows that consumed supplemental forage had a tendency for increased milk yield (38.03 vs. 35.95 kg, for F+ and F– respectively, SEM = 1.63, $P = 0.09$, Table 4). Cows with TMR access for 19 h/d had greater feed efficiency (Table 4) on both a milk volume (kg of milk/kg of DMI, $P = 0.03$) and component (kg of ECM/kg of DMI, $P = 0.05$) basis compared with the 24-h group. This improved efficiency is likely a result of longer feed retention times and improved total-tract digestibility. However, BW of cows did not change over the study. No differences were observed in milk protein, fat, and urea content; however, somatic cells tended to be higher for 19 h/d TMR diets, in particular when hay was not offered (Table 4). Lactose tended to be higher in milk from cows with 24-h TMR access, likely because of the lower SCC (Table 4).

Rumination times were much higher than values considered as a minimum threshold to ensure rumen functionality (>390 min/d; Zebeli et al., 2007, 2009). An interaction was observed between TMR availability and provision of supplemental hay for rumination time (Table 5). Cows with 19 h of TMR access ruminated less when hay was offered ($P < 0.01$). In contrast, hay had no effect on rumination time in cows with 24 h of

Table 3. Effect of feed restriction and supplemental hay¹ on fecal fiber content and total-tract fiber digestibility

Item	Feed availability				SEM	<i>P</i> -value	
	24 h		19 h			24 vs. 19 h	F+ vs. F−
	F+	F−	F+	F−			
Fecal aNDFom, ² % of DM	60.42	60.15	60.14	60.38	0.48	0.92	0.95
Fecal uNDFom, ³ % of DM	32.21	31.99	32.48	31.69	0.83	0.97	0.21
Fecal pdNDF, ⁴ % of DM	28.20	28.17	27.66	28.69	1.03	0.98	0.34
aNDFom TTD, ⁵ %	59.64	60.21	59.54	59.76	1.08	0.58	0.42
pdNDF TTD, %	75.90	76.30	76.10	75.69	1.37	0.76	0.99

¹F+ = supplemental hay; F– = no supplemental hay.

²Amylase- and sodium sulfite-treated NDF with ash correction.

³Unavailable NDF estimated via 240-h in vitro fermentation.

⁴Potentially digestible NDF.

⁵Total-tract fiber digestibility.

TMR access. Rumination time, average reticuloruminal pH values, and time when pH was below pH 5.8 and 5.5 are shown in Table 5. The results obtained are reflective of the high-fiber diets used in this study. Time in which the pH fell below 5.5, the minimum critical threshold, was negligible. Average pH values were lower ($P < 0.05$) in the diets where long hay was offered, but for no obvious reason. Overall pH of cows was not affected by diet treatment. This was likely a result of the cows' preference for TMR and their ability to somewhat compensate for total TMR DMI once adapted to the restricted hours of availability (Maulfair et al., 2013), while the group having no access to feed for 5 h ate less.

No differences were observed regarding average ammonia and ruminal VFA levels, as shown in Table 6. At 12 h after feeding (0800 h) a tendency was observed for lower ammonia for the cows with 19-h TMR access. This could be explained by the lower DMI for

the restricted diets that release a higher amount of fermentable carbohydrates, allowing bacteria to consume more nitrogen during the part of the day when feed was available. Conversely, when looking the values obtained 24 h after feed distribution (2000 h), acetate, propionate, and total VFA tended to be lower in cows with 19 h of TMR access, and butyrate was significantly lower. This condition is likely due to the absence of new feed that could have reduced VFA production in the rumen. All other rumen parameters measured were similar. Restricting TMR comprised of dry forages, whereas offering additional long hay appears to be consistent with previous research using silage-based diets (Kmicikewycz and Heinrichs, 2014). In the same research, once cows adapted to the restricted feeding period, the lack of hay made little change to their feed intake and digestion. This approach to feeding may also reduce nutrient excretion due to greater feed digestibility.

Table 4. Effect of feed restriction and supplemental hay¹ on milk production and quality

Item	Feed availability				SEM	P-value	
	24 h		19 h			24 vs. 19 h	F+ vs. F−
	F+	F−	F+	F−			
Milk, kg/d	37.87	36.56	38.19	35.33	1.63	0.71	0.09
ECM, kg/d	40.26	38.23	39.13	36.84	1.33	0.13	0.01
Milk kg/DMI kg	1.40	1.54	1.91	1.64	0.18	0.03	0.64
ECM kg/DMI kg	1.48	1.60	1.92	1.68	0.17	0.05	0.63
Fat, %	3.82	3.62	3.58	3.60	0.18	0.31	0.46
Fat yield, kg/d	1.27	1.32	1.35	1.33	0.16	0.71	0.91
Protein, %	3.38	3.38	3.42	3.31	0.11	0.74	0.34
Protein yield, kg/d	1.13	1.26	1.28	1.20	0.10	0.65	0.83
Lactose, %	4.88	4.88	4.80	4.79	0.05	0.04	0.94
Lactose yield, kg/d	1.65	1.83	1.86	1.75	0.15	0.68	0.85
Urea, %	14.68	15.42	15.30	15.53	1.36	0.72	0.65
SCC, log cfu/mL	2.39	3.49	3.38	4.00	0.80	0.07	0.04
BW variation, kg	0.52	−3.48	17.63	17.47	9.60	0.08	0.85

¹F+ = supplemental hay; F– = no supplemental hay.

Table 5. Effect of feed restriction and supplemental hay¹ on rumination time and reticuloruminal pH

Item	Feed availability				SEM	<i>P</i> -value	
	24 h		19 h			24 vs. 19 h	<i>P</i> + vs. <i>P</i> −
	<i>F</i> +	<i>F</i> −	<i>F</i> +	<i>F</i> −			
Rumination, min/d	424.98 ^a	443.10 ^a	446.26 ^a	412.22 ^b	30.49	0.62	0.41
Daily average pH	6.10	6.11	6.09	6.12	0.09	0.88	0.04
Time pH <5.8, min/d	18.79	19.06	22.02	18.44	11.78	0.32	0.21
Time pH <5.5, min/d	0.79	0.25	0.10	1.74	0.84	0.51	0.36

^{a,b}Means with different superscripts differ at $P < 0.05$ (significant interaction between effects of feed availability and provision of supplemental hay).

¹F+ = supplemental hay; F– = no supplemental hay.

CONCLUSIONS

The results obtained in this study show that once cows adapted to diet changes, few differences were observed when offering TMR continuously 24 h/d or limited to 19 h/d. In addition, the presence of long hay during the TMR restriction offered the best conditions for ruminal function and pH stability and tended to improve milk production. It was noted that a restriction of TMR availability for 5 h/d can potentially increase feed efficiency, as has been shown in other classes of ru-

minants. These results need to be confirmed by longer duration experiments using mid-lactation dairy cows. A Latin square with 21-d periods could have a carry-over effect on energy storage and mobilization of fat reserves might be able to mask negative energy balance during restriction. However, in our experiment we did not see any BW variation. This feeding system could be used as a strategy to manage feed availability according to cow production and metabolic condition and to maximize the use of nutrient resources, reducing the cost of milk production and improving the cows' welfare and health.

Table 6. Effect of feed restriction and supplemental hay¹ on ruminal VFA and ammonia content

Item	Feed availability				SEM	<i>P</i> -value	
	24 h		19 h			24 vs. 19 h	F+ vs. F–
	F+	F–	F+	F–			
Average, mmol/L							
NH ₃	3.95	4.00	3.33	3.56	0.49	0.24	0.75
Acetic	45.46	50.76	47.54	42.95	3.19	0.37	0.91
Propionic	18.94	21.92	20.26	18.29	1.86	0.48	0.76
Butyric	9.14	9.61	9.48	8.98	0.82	0.49	0.76
Isobutyric	0.47	0.56	0.49	0.45	0.08	0.83	0.98
Valerianic	0.91	1.07	1.06	0.95	0.09	0.88	0.84
Isovalerianic	0.92	1.02	1.10	0.98	0.13	0.50	0.96
Total VFA	75.84	84.94	79.93	72.60	5.14	0.45	0.87
12 h after feeding, mmol/L							
NH ₃	4.23	4.70	3.54	3.24	0.57	0.07	0.88
Acetic	39.77	48.38	47.03	42.78	4.37	0.86	0.63
Propionic	15.11	19.63	20.13	17.71	2.09	0.46	0.62
Butyric	7.77	8.96	9.95	9.22	1.11	0.24	0.82
Isobutyric	0.42	0.61	0.45	0.40	0.09	0.34	0.42
Valerianic	0.79	0.99	1.05	0.95	0.12	0.37	0.70
Isovalerianic	0.76	1.03	0.97	0.96	0.15	0.56	0.30
Total VFA	64.62	79.59	79.59	72.03	7.18	0.62	0.62
24 h after feeding, mmol/L							
NH ₃	3.66	3.31	3.11	3.87	0.51	0.99	0.68
Acetic	51.17	53.17	47.99	42.61	3.86	0.10	0.69
Propionic	22.76	24.20	20.42	19.07	2.45	0.10	0.96
Butyric	10.53	10.28	8.97	8.40	0.82	0.03	0.56
Isobutyric	0.53	0.51	0.53	0.48	0.08	0.81	0.59
Valerianic	1.04	1.14	1.07	0.92	0.11	0.44	0.89
Isovalerianic	1.07	1.01	1.21	0.92	0.14	0.79	0.16
Total VFA	87.08	90.30	80.22	72.75	6.35	0.09	0.77

¹F+ = supplemental hay; F– = no supplemental hay.

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