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Modulation of rumen pH by sodium bicarbonate and a blend of different sources of magnesium oxide in lactating dairy cows submitted to a concentrate challenge

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ABSTRACT

With the objective of evaluating the potential effects of sodium bicarbonate or a magnesium-based product on rumen pH and milk performance of dairy cattle exposed to a dietary challenge, 30 lactating Holstein cows $(648 \pm 67 \text{ kg of body weight}; 44.4 \pm 9.9 \text{ kg/d of milk})$ yield; 155 ± 75 d in milk) were blocked by parity (9 primiparous and 21 multiparous) and randomly distributed to 3 treatment groups. One group received a total mixed ration (TMR) that acted as a control (CTR), a second group (SB) received the same TMR but with an additional supplementation of 0.8% of sodium bicarbonate, and a third group (MG) received the same TMR as CTR but an additional supplementation of 0.4% of a magnesium-based product (pHix-Up, Timab, Dinard, France). After 1 wk of exposure to this TMR, all 3 rations were supplemented with 1 kg/d of barley, which was then increased 1 kg/wk until reaching 3 kg/d of barley during wk 4 of the study. Every kilogram of barley replaced 1 kg of forage in the diet. Individual feed intake and behavior were monitored using electronic feed bins. Seven cows per treatment were equipped with an intraruminal bolus that recorded pH every 15 min. As the severity of the barley challenge increased, dry matter intake decreased, but this decrease was more pronounced in SB cows than in MG cows, with an intermediate response for CTR cows. The MG cows produced more milk when challenged with 2 or 3 kg/d of additional barley than when challenged with 1 kg/d, whereas CTR cows produced less milk with the 3 kg/d challenge compared with 1 or 2 kg/d, and the SB cows maintained milk production. Milk fat content decreased with barley challenges, with CTR cows experiencing a more severe decrease than SB cows, which maintained stable butterfat values throughout the study, and MG cows showed a decline in milk fat content only with the 3 kg/d of additional barley. Meal size was also reduced as the severity of barley challenge increased, and this reduction was more modest in MG cows than in SB cows. The number of daily meals consumed by SB and MG cows was more constant than that recorded in CTR cows. Cows on the CTR and SB treatments showed a marked decrease in rumen pH with the 3 kg/d of additional barley, whereas MG cows maintained stable rumen pH during the barley challenges and had greater average rumen pH (5.93 ± 0.04) than CTR cows (5.83) \pm 0.04) with the 3 kg/d of additional barley; SB cows showed intermediate values (5.85 \pm 0.04). Last, MG cows spent less time $(32.3 \pm 6.1\%)$ with rumen pH <5.8 when exposed to the 3 kg/d of barley challenge than CTR and SB cows ($50.7 \pm 5.02\%$). In conclusion, supplementation with MG prevents the decline in dry matter intake and milk production induced by a rumen challenge, whereas supplementation with SB prevents the decay in milk production but does not prevent the decrease in feed intake. These changes were probably due to the ability of the MG treatment to prevent a reduction in rumen pH when challenging cows with 3 kg/d of additional barley in the ration.

Key words: fermentation, neutralization, rumen acidosis

INTRODUCTION

Milk production in dairy cattle has progressively increased throughout the years, and thus energy density of the ration has been increasing in an attempt to meet the nutrient demands of cows. The increase in energy density has been achieved though incremental amounts of NFC and to some extent by adding fat to the diet. However, increasing the amount of NFC may lead to more vigorous rumen fermentation and accumulation of fermentation end products (mainly VFA, but also

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on occasion lactic acid), which may decrease rumen pH (Agle et al., 2010; Blanch et al., 2010). The reduction of rumen pH may alter rumen microbiota (Petri et al., 2013) and cause some dysfunction, such as alterations in feeding and rumination patterns and total feed intake (DeVries et al., 2009) and reductions in milk fat content (Rustomo et al., 2006). With the aim of controlling rumen pH, many studies have evaluated the use of supplementing rations with buffers or alkalinizers (or neutralizing agents) such as sodium bicarbonate or magnesium oxide, respectively, as well as supplementing live yeast (Thrune et al., 2009; DeVries and Chevaux, 2014) and yeast cultures (Poppy et al., 2012). Most studies have reported increases in DMI (Erdman et al., 1982; Rogers et al., 1985a; Staples et al., 1988), milk yield (Kilmer et al., 1981; Thomas et al., 1984; Rogers et al., 1985a), and milk fat content (Rogers et al., 1985a; Solorzano et al., 1989) when cows were supplemented with sodium bicarbonate, although some (Donker and Marx, 1980; DePeters et al., 1984; Rogers et al., 1985b) found no responses. Whether changes were directly caused by sodium bicarbonate or by an increase in DCAD is unknown, but several authors have reported improvements in milk yield and DMI when DCAD is increased (Sanchez and Beede, 1996; Hu and Murphy, 2004; Iwaniuk and Erdman, 2015). Less information is available about the effect of magnesium oxide on performance of dairy cows (Emery et al., 1965; Xin et al., 1989; Tebbe et al., 2018) and feed intake (Beede, 2017). The effectiveness of magnesium oxide sources in raising rumen pH and fostering improvements in milk yield and milk fat has been reported to differ across sources (Schaefer et al., 1982; Leno et al., 2017; Tebbe et al., 2018), and Lough et al. (1990) showed that magnesium oxide was more effective than magnesium chelate in promoting increases in DMI, milk yield, and milk fat content. The acid-neutralizing capacity of sodium bicarbonate and especially magnesium oxide depends on several physical and chemical characteristics, which lead to different rates of solubilization in the rumen fluid (Le Ruyet and Tucker, 1992). Magnesium sources (typically carbonates) are calcined at different temperatures and over different exposure times (ranging from minutes to days) to obtain magnesium oxide, and both temperature and particle size have an effect on their properties. For instance, low calcination temperatures ($<500^{\circ}$ C) generate magnesium oxides that have a porous structure with a large surface area and increased reactivity (Eubank, 1951). The objective of this study was to evaluate the effects of sodium bicarbonate or a magnesium-based product on rumen pH and milk performance of dairy cattle exposed to a dietary challenge aimed at inducing a decrease in rumen pH.

MATERIALS AND METHODS

Animals and Treatments

All procedures described herein were supervised and approved by the Institut de Recerca i Tecnologia Agroalimentàries Animal Care Committee (Barcelona, Spain). Thirty lactating Holstein cows (648 \pm 66.6 kg of BW; $44.4 \pm 9.9 \text{ kg/d}$ of milk yield; $155 \pm 75 \text{ DIM}$; mean \pm SD) were blocked by parity (9 primiparous and 21 multiparous) and randomly distributed to 3 treatment groups. Cows were housed in a single pen equipped with freestalls bedded every 2 d with chopped straw and electronic feed bins (MooFeeder, MooSystems, Cortes, Spain) that controlled the access of different cows in the same pen to specific bins containing the different dietary treatments. The electronic bins, in addition to controlling access to feed, recorded the time of day and amount of feed consumed at every visit throughout the study.

One group of cows received a TMR that met current nutrition recommendations (NRC, 2001) and acted as a control (**CTR**), a second group (**SB**) received the same TMR but with an additional supplementation of 0.8%(DM basis) of sodium bicarbonate (Solvay Química SL, Barcelona, Spain) to supply an expected amount of 200 g/d, and a third group (MG) received the same TMR as CTR but supplemented with 0.4% (DM basis) of a magnesium-based product [pHix-Up, Timab, Dinard, France; a blend of magnesium oxide (81%), calcium oxide (5.5%), and other mineral components naturally present in the original ores obtained from different calcination processes involving temperatures ranging between 600 and 1,500°C] to supply an expected amount of 100 g/d. The reactivity of the magnesium-based product in citric acid ranged from 140 to 170 min. The product contained 48.5% elemental magnesium with a high alkalinizing capacity ($\sim 38 \text{ mEq/g}$) and different solubilization kinetics providing a fast-acting and longlasting effect (inducing in an in vitro rumen simulation medium an increase of pH from 5.5 to 5.8 in less than 2 h, which was then maintained for at least 6 h). The dose of 200 g/d (or 0.8%) of sodium bicarbonate was based on the conclusions from the meta-analysis conducted by Hu and Murphy (2005), who reported that supplementation between 0.7 and 1% of DM as sodium bicarbonate was optimal for early- and mid-lactation cows; the MG dose was then set at half that of sodium bicarbonate based on its expected neutralizing activity. Using a half dose of magnesium relative to sodium bicarbonate was based on a preliminary study to compare the neutralizing capacity of magnesium relative to sodium bicarbonate. The neutralizing capacity was assessed

Wk 4 Wk 1 Wk 2 Wk 3 CTR MG MGCTR MG Item SB CTR SBSB CTR SB MG Ingredient, % of DM Corn silage 4.94.94.94.94.94.94.94.95.15.15.14.9Barlev 20.420.220.325.325.125.228.228.028.132.131.932.0 Grass silage 5.25.25.05.05.05.25.05.05.05.05.05.013.413.39.3 9.3Fescue hav 14.714.614.613.411.1 11.1 11.1 9.3 Corn 19.819.719.820.720.520.619.919.719.819.919.719.89.49.39.46.0 6.0 4.6Rve grass hav 4.04.04.06.04.54.610.2Soybean meal 9.99.89.8 10.310.29.9 9.89.9 9.99.89.8Straw 3.13.13.12.62.62.62.02.02.01.51.51.52.22.12.12.12.12.12.1Alfalfa hav 2.1 2.12.12.12.1 Sunflower meal 3.93.93.94.14.14.13.93.93.9 3.9 3.93.9Soybean hulls 1.71.71.71.71.71.71.71.71.71.71.71.7Molasses 1.00.90.91.01.01.01.00.91.01.00.91.0Carob flour 1.1 1.1 1.1 1.21.2 1.21.1 1.11.11.1 1.1 1.1 0.90.90.90.80.8Palm oil 0.80.80.80.80.80.8 0.8 Calcium carbonate 1.01.01.01.01.01.01.01.01.01.01.01.00.5Beet pulp 0.50.50.50.50.50.50.50.50.50.50.5Salt 0.40.40.40.40.40.40.40.40.40.40.40.4Dicalcium phosphate 0.10.10.10.10.10.10.10.10.10.10.10.10.1Urea 0.10.10.10.10.10.10.10.10.10.10.1Vitamin-mineral premix² 0.20.20.20.20.20.20.20.20.20.20.20.20.0 0.8 0.0 0.0 0.8 0.0 0.0 0.8 0.0 0.0 0.8 0.0 Sodium bicarbonate pHixUp 0.0 0.0 0.40.00.00.40.0 0.00.40.0 0.00.4Nutrient NE_L , Mcal/kg1.631.571.571.651.591.591.671.601.601.691.621.62CP, % 14.914.314.315.114.514.515.314.714.715.514.914.9NFC. % 42.646.244.344.347.945.949.647.544.542.645.947.5NDF, % 31.3 30.0 30.029.528.328.327.826.626.626.024.924.9Fat, % 2.22.12.12.22.12.12.32.22.22.32.22.2Ash, %6.67.16.86.86.96.66.66.86.56.56.36.3Ca, % 0.760.730.770.740.730.750.720.740.750.710.700.71P, % 0.410.420.420.420.420.400.400.400.430.410.410.40Mg, %0.20 0.210.39 0.190.20 0.370.22 0.20 0.36 0.190.170.37Cl, % 0.560.580.470.540.510.470.480.450.460.520.550.56K, % 1.541.521.521.481.461.421.361.411.431.171.141.17Na, % 0.290.470.360.36 0.530.400.430.580.400.400.610.39 S, % 0.190.190.180.190.190.190.180.190.190.160.160.17DCAD, mEq/100 g24.726.727.128.228.822.531.535.136.629.030.020.3

Table 1. Ingredient and nutrient composition of the TMR fed during the study¹

 1 CTR = control ration (no supplementation); SB = control ration supplemented with 0.8% of sodium bicarbonate; MG = control ration supplemented with 0.4% of a magnesium-based product (pHix-Up, Timab, Dinard, France).

 2 Contained 81.6 mg/kg of Zn; 11.5 mg/kg of Cu; 57.6 mg/kg of Mn; 9.86 mg/kg of Co; 1.92 mg/kg of I; 0.34 mg/kg of Se; 58 mg/kg of S; 120,000 IU/kg of vitamin A; 28,800 IU/kg of vitamin D; and 1,920 IU/kg of vitamin E.

as the milliequivalents of protons (from hydrochloric acid) required to lower the pH of a solution containing 2.5 g of magnesium or sodium bicarbonate in a volume of 50 mL to a pH of 3 starting from a pH of either 5.5 or 6.5. At both pH 5.5 and pH 6.5, the neutralizing capacity of sodium bicarbonate was approximately 12 mEq/g, whereas that of magnesium was approximately 38 mEq/g.

After 1 wk of exposure to this TMR, the ration for all treatment groups was supplemented with 1 kg/d of barley. The amount of additional barley was increased by 1 kg/wk until reaching 3 kg/d of barley in wk 4 of the study. Thus, the study lasted 4 wk. Every kilogram of barley replaced 1 kg of forage (fescue hay and ryegrass hay). The ingredient and nutrient compositions of the

different TMR used in the study are depicted in Table 1. Cows were fed twice daily (at 0700 and 1600 h). All cows had free access to water and were fed ad libitum.

Individual feed intake and feeding behavior were monitored using electronic bins (Bach et al., 2004). Individual milk production at every milking was determined using electronic milk meters (AfiMilk, Afikim Ltd., Kibbutz Afikim, Israel), and milk fat and milk protein contents were determined electronically every milking using the AfiLab system (Afikim Ltd.), which was calibrated every 2 wk. Similarly, lying time was monitored and recorded on a daily basis using pedometers (Afikim Ltd.)

The TMR was sampled every 2 d, and samples were composited by week and analyzed for DM (method

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934.01), ash (method 942.05), ether extract (method 920.39), and N (method 984.13) content following AOAC International (2000) and for NDF according to Van Soest et al. (1991) using an Ankom220 Fiber Analyzer unit (Ankom Technology Corp., Fairport, NY) using sodium sulfite and a heat-stable amylase. Nonfiber carbohydrates were calculated as 100 minus CP, NDF, ether extract, and ash. Also, TMR samples were analyzed for Ca, P, Na, K, Cl, S, and Mg using inductively coupled plasma MS. At the beginning of the study, 7 cows per treatment were equipped with an intraruminal bolus (eBolus, eCow Devon Ltd., Devon, UK) that recorded pH every 15 min.

Calculations and Statistical Analyses

Energy-corrected milk was calculated following NRC (2001) as

ECM $(kg/d) = 12.55 \times fat yield (kg/d) + 7.39$

 \times protein yield (kg/d) + 5.34 \times lactose yield (kg/d).

Dietary cation-anion difference was calculated following Jackson et al. (2001) as

DCAD (mEq/100 g) = {[Na (g/kg)/0.0023]
+ [K (g/kg)/0.00391]} - {[Cl (g/kg)/0.00355]
+ [S (g/kg)/0.00321]
$$\times$$
 2}.

All data were checked for normality before conducting the mixed-effects model and were transformed to a normal distribution when original data did not follow a normal distribution (according to the Shapiro-Wilk test). The mixed-effects model included the fixed effects of treatment (CTR, SB, or MG), time of sampling, challenge (1, 2, or 3 kg/d of additional barley), and their 2- and 3-way interactions plus the random effect of cow and block (parity). Time entered the model as a repeated measure using the unstructured, compound symmetry, and autoregressive order 1 covariate-variance matrices, and the one with the lowest Bayesian criterion was selected. Average values for the 7 d without barley challenge (wk 1) were used as a covariate in the model. All data presented herein are adjusted values using this model.

RESULTS AND DISCUSSION

DMI and Milk Production and Composition

Dry matter intake was greater (P < 0.01) in MG than in SB cows, with CTR cows having intermediate

values (Table 2). As the severity of the barley challenge increased, feed intake decreased, but this decrease was more pronounced (P < 0.01) in SB than in MG cows, with an intermediate response for CTR cows (Figure 1). Several studies have reported that rumen acidosis causes a decrease in DMI (Owens et al., 1998). Thus, the maintenance of DMI observed herein in MG cows exposed to 3 kg/d of additional barley compared with SB cows could be associated with improved rumen pH conditions. However, previous studies have reported no changes (Erdman et al., 1982; Rauch et al., 2012) or decreases (Thomas et al., 1984) in DMI when supplementing cows at risk of rumen acidosis with magnesium-derived products, and Beede (2017) reported a 3% decline in DMI when increasing the inclusion of magnesium oxide in the diet from 0.21 to 0.46%. Nevertheless, there is a great divergence in the quality and characteristics of magnesium oxide products (Beede, 2017), and thus the different results obtained herein compared with previous reports could be partly explained by the intrinsic characteristics of the different sources of magnesium oxide. Furthermore, there was an interaction between day and barley challenge mainly due to the fact that feed intake was steadier during the 7 d of challenge with the 2 kg of additional barley than with the 1 and 3 kg of additional barley regardless of treatment, with cows on 3 kg of additional barley showing the most fluctuation in DMI (Figure 2). Previous literature has commonly referred to fluctuations in DMI when both beef (Britton and Stock, 1986; Enemark, 2008) and dairy (Nocek et al., 2002) cattle undergo rumen acidosis. In the current study, the coefficient of variation in DMI between days with the 1 and 3 kg/d of additional barley (22.5 and 24.7%) was greater than that with the 2 kg/d of additional barley (17.0%), with the lowest (P < 0.05) coefficient of variation of DMI observed for the base period when no barley challenge was applied (12.0%).

Overall, there were no differences in milk production (Table 2), but there was an interaction (P < 0.05) between treatment and severity of concentrate challenge: MG cows produced more milk when challenged with 2 or 3 kg/d of additional barley than when challenged with 1 kg/d, whereas CTR cows produced less milk with the 3 kg/d challenge compared with 1 or 2 kg/d, and SB cows maintained steady milk production throughout the study (Figure 3). Performance of CTR cows followed a similar pattern for DMI, producing less milk when DMI was reduced with 3 kg/d of additional barley. Cows on the MG treatment produced more milk with 2 and 3 kg of additional barley, and SB cows maintained steady milk production throughout the different barley challenges despite the fact that DMI decreased

Table 2. Performance and behavior of dairy cows as affected by pH-neutralizing additives and inclusion of barley in the diet

	$Treatment^1$				P-value ²						
Item	CTR	SB	MG	SEM	Т	С	D	$\mathbf{T}\times\mathbf{C}$	$T \times D$	$C \times D$	$T \times C \times D$
DMI, kg/d	22.1 ^{ab}	21.0 ^b	23.7^{a}	1.28	< 0.01	< 0.01	< 0.01	0.03	0.58	< 0.01	0.23
Milk, kg/d	40.4	39.6	40.3	0.79	0.76	0.20	0.37	0.02	0.89	0.53	0.36
Fat, %	3.50	3.57	3.54	0.05	0.33	< 0.001	0.08	< 0.01	0.51	< 0.01	0.66
Protein, %	3.46	3.47	3.45	0.04	0.45	0.04	0.20	0.50	0.49	0.08	0.07
ECM, kg/d	41.3	40.9	41.4	0.77	0.88	< 0.01	0.88	< 0.01	0.91	0.52	0.09
Lying time, min/d	677	694	674	42.7	0.94	< 0.01	< 0.01	0.94	0.49	< 0.01	0.76
Meal size, kg	4.53	4.25	4.68	0.18	0.25	< 0.01	< 0.01	0.01	0.10	< 0.01	0.51
Meal interval, min	217	213	205	14.2	0.87	0.04	0.03	0.86	0.54	< 0.01	0.44
Meals, no./d	6.28	6.41	6.08	0.20	0.56	< 0.01	< 0.01	$<\!0.01$	0.77	$<\!0.01$	0.99

^{a,b}Values with different superscripts within a row differ (P < 0.05).

 1 CTR = control ration (no supplementation); SB = control ration supplemented with 0.8% of sodium bicarbonate; MG = control ration supplemented with 0.4% of a magnesium-based product (pHix-Up, Timab, Dinard, France).

 $^{2}T = \text{effect of treatment; } C = \text{effect of challenge (or week); } D = \text{effect of day within challenge.}$

with 3 kg/d of additional barley in this group of animals.

Milk fat content decreased (P < 0.05), irrespective of treatment, from 3.59 to 3.54%, ending in 3.46% with challenges of 1, 2, and 3 kg/d of additional barley, respectively. Furthermore, milk fat content evolved differently (P < 0.01) across barley challenges depending on treatment: CTR cows experienced a more severe decrease in milk fat content than SB cows, which maintained stable butterfat values throughout the study (Figure 4), and MG cows showed a decline in milk fat content with 3 kg/d of additional barley (Figure 4). Furthermore, these differences were more marked (P < 0.01) as the days on barley challenge increased (with final values on d 7 of 3.56, 3.50, and 3.43% for 1, 2, and 3 kg/d of barley, respectively) regardless of treatment. On the other hand, milk protein content was reduced (P < 0.05) from 3.48% with 1 kg/d to 3.463% with 2 and 3 kg/d of additional barley independent of treatment.



Figure 1. Dry matter intake (kg/d) of cows exposed to a challenge of 1, 2, and 3 kg/d of additional barley in the ration along with no supplementation (control; CTR) or supplementation of 0.8% of sodium bicarbonate (SB) or 0.4% of a magnesium oxide-based product (MG; pHix-Up, Timab, Dinard, France). Error bars depict SEM (not computed for the baseline period). Columns with different lowercase letters (a, b) within treatment differ (P < 0.05). Columns with different uppercase letters (A–C) between treatments differ (P < 0.05).



Figure 2. Dry matter intake (kg/d) during a 7-d challenge of 1, 2, and 3 kg/d of additional barley in the ration. Bars depict SEM. Values with different letters (a, b) within time points differ (P < 0.05).

different milk components, ECM was calculated. The production of ECM was similar with 1 and 2 kg/d of production in CTR cows, as SB and MG cows main-

To account for the combination of milk yield and additional barley (41.4 kg/d), but it decreased (P <0.05) with 3 kg/d (40.8 kg/d), mainly due to a reduced



Figure 3. Milk production (kg/d) of cows exposed to a challenge of 1, 2, and 3 kg/d of additional barley in the ration along with no supplementation (control; CTR) or supplementation of 0.8% of sodium bicarbonate (SB) or 0.4% of a magnesium oxide-based product (MG; pHix-Up, Timab, Dinard, France). Error bars depict SEM (not computed for the baseline period). Columns with different letters (a, b) within treatment differ (P < 0.05).

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Figure 4. Milk fat content (%) of cows exposed to a challenge of 1, 2, and 3 kg/d of additional barley in the ration along with no supplementation (control; CTR) or supplementation of 0.8% of sodium bicarbonate (SB) or 0.4% of a magnesium oxide-based product (MG; pHix-Up, Timab, Dinard, France). Error bars depict SEM (not computed for the baseline period). Columns with different letters (a–c) within treatment differ (P < 0.05).



Figure 5. Production of ECM of cows exposed to a challenge of 1, 2, and 3 kg/d of additional barley in the ration along with no supplementation (control; CTR) or supplementation of 0.8% of sodium bicarbonate (SB) or 0.4% of a magnesium oxide-based product (MG; pHix-Up, Timab, Dinard, France). Error bars depict SEM (not computed for the baseline period). Columns with different letters (a, b) within treatment differ (P < 0.05).

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tained ECM production even with the 3 kg/d of barley challenge (Figure 5).

The amount of time that cows spent lying down was greatest (P < 0.01) with 2 kg/d of additional barley (698 min/d) followed by 3 kg/d (681 min/d) and 1 kg/d (667 min/d) of additional barley. Furthermore, as days exposed to a barley challenge increased, lying time progressively decreased from 720 min on d 1 to 612 min on d 7 of each challenge period; this decrease was more marked with 1 and 3 kg/d than with 2 kg/d of additional barley (Figure 6), independent of dietary treatments. These results are in line with previous reports describing a reduction in lying time in cows exposed to rumen acidosis (DeVries et al., 2009).

Feeding behavior was also affected by the addition of barley in the ration. As the amount of additional barley increased (P < 0.01), meal size was reduced from 4.80 kg/meal with 1 kg/d of barley to 4.44 or 4.22 kg/meal with 2 and 3 kg/d of barley, respectively, and this decrease was more (P < 0.01) pronounced toward the end of each 7-d period than during the beginning of exposure to the barley challenge (with an average meal size of 4.88 kg/d), independent of treatment (with final values on d 7 of 4.43, 4.17, and 3.91 g/min for 1, 2, and 3 kg/d of barley, respectively). Furthermore, SB cows had a marked (P < 0.01) decrease in meal size (Figure 7) when addition of barley increased from 1 to 2 or 3 kg/d (from 4.84 kg/meal to 4.06 and 3.86 kg/ meal, respectively). Similarly, CTR cows experienced a reduction of meal size from 4.75 kg/meal with 1 kg/d of additional barley to 4.31 kg/meal with 3 kg/d of additional barley (Figure 7). In contrast, meal size in MG cows was reduced (Figure 7) with 3 kg/d of additional barley (4.29 kg/meal) compared with 1 or 2 kg/d of additional barley (4.88 kg/meal). Furthermore, meal interval increased (P < 0.05) from 198 min with 1 kg/d of additional barley to 221 min with 2 and 3 kg/d of additional barley, and this decrease was more marked (P < 0.01) as days of exposure to barley challenge increased, especially with the 3 kg/d of additional barley, which increased from 211 min at d 1 to 293 min at d 7. Last, the number of daily meals was also reduced (P < 0.01) when comparing 1 or 2 kg/d of barley (6.56 meals/d) with 3 kg/d of additional barley (5.55 meals/d), and it was affected by an interaction between treatment and barley challenge, with SB and MG cows showing a more steady maintenance of number of meals (decreasing from 6.32 and 6.33 meals/d to 5.60 and 5.62 meals/d, respectively), whereas CTR cows experienced a much larger decrease in the number of meals (from 6.72 to 5.43 meals/d). DeVries et al. (2009) also reported a reduction in meal size and meal frequency following a rumen acidosis challenge, and it has been previously described that rumen acidosis results in longer meal intervals (Bach et al., 2007; DeVries and Chevaux, 2014). Nevertheless, the fact that meal size and meal frequency were more stable in MG cows (where meal size decreased only with 3 kg/d of barley)



Figure 6. Lying time (h/d) during a 7-d challenge of 1, 2, and 3 kg/d of additional barley in the ration. Bars depict SEM. Values with different letters (a, b) within time points differ (P < 0.05).

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Figure 7. Meal size (kg/meal) of cows exposed to a challenge of 1, 2, and 3 kg/d of additional barley in the ration along with no supplementation (control; CTR) or supplementation of 0.8% of sodium bicarbonate (SB) or 0.4% of a magnesium oxide-based product (MG; pHix-Up, Timab, Dinard, France). Error bars depict SEM (not computed for the baseline period). Values with different letters (a, b) within treatment differ (P < 0.05).

might indicate a better rumen environment supported, as described below, by an increased rumen pH.

Rumen pH

A pH meter failed for 1 cow in the MG treatment, and all data pertaining to this cow were removed from the statistical analysis. Average rumen pH during the baseline period was 6.07, and as expected it decreased (P < 0.05) to 5.93 and 5.94 with challenges of 1 and 2 kg/d of additional barley, respectively, and to 5.87 with 3 kg/d of additional barley. Overall, there were no changes in rumen pH due to treatment (Table 3), but there was an interaction (P < 0.05) between treatment and barley challenge (Figure 8). Cows on the CTR and SB treatments experienced a marked decrease in rumen pH with 3 kg/d of additional barley, whereas MG cows maintained stable rumen pH during the 3 barley challenges, resulting in greater (P < 0.05) rumen pH in MG cows (5.93) than in CTR cows (5.83) with the 3 kg/d of additional barley, with SB cows showing intermediate values (5.85). Furthermore, rumen pH progressively decreased as days exposed to the barley challenges increased, independent of treatment (Table 3). The proportion of time that rumen pH was ≤ 5.8 was not affected, overall, by treatment (Table 3), but as the addition of barley increased, the proportion of time with pH ≤ 5.8 increased (P < 0.01) from 37.5% (9 h/d) with 1 kg/d of additional barley to 44.6% (10.7 h/d) with 3 kg of additional barley. Furthermore, there was an interaction between treatment and barley challenge (Table 3). The MG cows spent less time with rumen pH \leq 5.8 when exposed to 3 kg/d of barley challenge than the SB and CTR cows (Figure 8). Also, CTR and

Table 3. Rumen pH in lactating cows as affected by treatment

$\mathrm{Treatment}^1$						P-value ²						
Item	CTR	SB	MG	SEM	Т	С	D	$\mathbf{T}\times\mathbf{C}$	$\mathbf{T}\times\mathbf{D}$	$C \times D$	$T \times C \times D$	
Rumen pH Time pH ≤ 5.8 , %	$5.91 \\ 45.8$	$5.92 \\ 37.7$	$5.93 \\ 37.5$	$\begin{array}{c} 0.03 \\ 5.99 \end{array}$	$0.86 \\ 0.52$	$< 0.01 \\ < 0.01$	<0.01 <0.01	<0.01 <0.01	0.20 0.77	<0.01 0.09	$0.81 \\ 0.52$	

 1 CTR = control ration (no supplementation); SB = control ration supplemented with 0.8% of sodium bicarbonate; MG = control ration supplemented with 0.4% of a magnesium-based product (pHix-Up, Timab, Dinard, France).

 $^{2}T = \text{effect of treatment; } C = \text{effect of challenge (or week); } D = \text{effect of day within challenge.}$



Figure 8. Average rumen pH of cows exposed to a challenge of 1, 2, and 3 kg/d of additional barley in the ration along with no supplementation (control; CTR) or supplementation of 0.8% of sodium bicarbonate (SB) or 0.4% of a magnesium oxide-based product (MG; pHix-Up, Timab, Dinard, France). Error bars depict SEM (not computed for the baseline period). Columns with different lowercase letters (a, b) within treatment differ (P < 0.05). Columns with different uppercase letters (A, B) between treatments differ (P < 0.05).



Figure 9. Proportion of observations with rumen pH \leq 5.8 of cows exposed to a challenge of 1, 2, and 3 kg/d of additional barley in the ration along with no supplementation (control; CTR) or supplementation of 0.8% of sodium bicarbonate (SB) or 0.4% of a magnesium oxide-based product (MG; pHix-Up, Timab, Dinard, France). Error bars depict SEM (not computed for the baseline period). Columns with different lowercase letters (a, b) within treatment differ (P < 0.05). Columns with different uppercase letters (A, B) between treatments differ (P < 0.05).

SB cows experienced an increase in the proportion of time with rumen pH \leq 5.8 when comparing 1 and 3 kg/d of additional barley; however, MG cows actually experienced a decrease in the proportion of time with rumen pH \leq 5.8 (Figure 9). These results indicate that MG cows were able to neutralize rumen pH when challenged with increasing doses of barley more efficiently compared with SB and CTR cows.

CONCLUSIONS

Rumen acidosis in dairy cattle results in increased fluctuations in total daily DMI and a progressive decrease in feed intake. However, supplementation with the magnesium-based product used herein prevents the decline in feed intake and milk production, whereas supplementation with sodium bicarbonate prevents the decay in milk production but does not prevent the decrease in feed intake. These changes were most likely due to the ability of the magnesium-based product used herein to prevent a reduction in rumen pH when challenging cows with 3 kg/d of additional barley in the ration.

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