Effects of two new formulas of dietary buffers with a high buffering capacity containing Na or K on performance and metabolism of mid-lactation dairy cows

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ABSTRACT

The present study was conducted to evaluate the effect of two new formulas of dietary buffers on intake, total tract digestibility, rumen pH, blood metabolites, and milk production of mid-lactation dairy cows. Nine multiparous cows (594 $\pm$ 46 kg BW; mean $\pm$ SD) averaging 120 $\pm$ 28 days in milk and producing 46.6 $\pm$ 3.4 kg/d were randomly assigned to a triplicate 3 × 3 Latin square. During each 21-d period, cows were offered one of three total mixed rations that varied in dietary buffer. The three types of dietary buffer were 1) 11.2 g/kg of dietary dry matter (DM) sodium bicarbonate (SB; control), 2) 8.7 g/kg of dietary DM high buffering capacity formula contained Na (HBNa), and 3) 7.4 g/kg of dietary DM high buffering capacity formula contained K (HBK). Each period was comprised of 14 d of dietary adaptation followed by 7 d of sampling. Measured buffering capacity was 102, 150 and 137 percent of $NaHCO_3$ for SB, HBNa and HBK, respectively. The amount of Na and K were 270 and 0, 310 and 0, and 250 and 60 g/kg for SB, HBNa, and HBK, respectively. Dry matter intake (DMI) tended ($P = 0.06$) to be lower with HBK (20.6 kg/d) than SB (21.0 kg/d) and HBNa (21.2 kg/d). No treatment effects were observed on rumen pH (averaged 5.88) and DM digestibility in the total digestive tract (averaged 79.4%). Yields of actual milk (38.1 kg/d) and 3.5% fat corrected milk (31.6 kg/d) were not affected by treatments, whereas yields of solid corrected milk ($P = 0.07$) and milk fat ($P = 0.10$) tended to be greater with HBK than SB and HBNa. Milk fat concentration in cows fed HBK was greater than in cows fed other treatments (32.5 vs. 29.5 and 29.6 g/kg; $P = 0.04$). Concentration of milk protein (32.2 vs. 30.6 g/kg) and lactose (46.8 vs. 44.4 g/kg) also were greater in cows fed HBK than those fed SB ($P = 0.02$). Efficiency of milk production was greater in cows fed HBK than SB (1.86 vs. 1.80; $P = 0.01$), whereas efficiency of solid corrected milk production was greater in HBK than SB and HBNa (1.64 vs. 1.51 and 151; $P = 0.02$). Blood concentration of Ca was higher with HBK compared with SB and HBNa (10.4 vs. 9.7 and 9.9 mg/dL, respectively; $P = 0.01$). These results indicated that under the current experimental condition, supplementation of dairy cow diet with a high buffering capacity buffer containing 60 g/kg K decreased DMI and improved milk composition and milk efficiency of mid-lactation dairy cows.

1. Introduction

Obtaining buffer and electrolyte balance requirements of dairy cows has been a prolonged objective of ruminant nutritionists and researchers because of the improving effects on cow heath and production (Erdman, 1988; Iwaniuk and Erdman, 2015). Several modes of action have been attributed to supplemental dietary buffers, including modification of ruminal pH, blood pH, electrolyte balance, rumen microbial synthesis, bioactive intermediates of ruminal fatty acid biohydrogenation, and reactions to environmental stressors (Sanchez et al., 1994;

Shire and Beede, 2013; Iwaniuk and Erdman, 2015). Despite considerable efforts, rumen acidosis and milk fat depression remain as common problems of dairy cows (Esmaeili et al., 2016; Oetzel, 2017), especially during of metabolic (Nasrollahi et al., 2017b) or heat (Staples and Thatcher, 2011) stresses. Although individual variability among animals can partly describe the phenomenon (Nasrollahi et al., 2017a), the buffering requirements of dairy cows have not been completely characterized and are likely deficient in many diets (Humer et al., 2018).

Increasing the specific buffering capacity of the dietary buffer is one

potential way of obtaining increased total buffering capacity in a dairy cow diet. A recent study reported that increasing buffering capacity improved rumen fermentation (Wu, et al., 2015). Data from a wide range of dry feeds suggest that total ash and cation contents are good indicators of total buffering capacity (Jasaitis et al., 1987). Mineral solubility also is considered to be an important factor for efficacy of additive buffers, as common buffers vary considerably in acid neutralizing capacity and solubility (Erdman, 1988).

Other than buffering capacity, the mineral cation of supplemental buffers has been shown to have specific effects on cow health and production (West et al., 1986; Iwaniuk et al., 2015; Alfonso-Avila et al., 2017). Indeed, Na and K, which are common mineral cations in additive buffers, have been shown to affect rumen and blood pH, acid base status, rumen biohyrogenation, and milk composition independently of buffering capacity and dietary cation-anion difference (DCAD) (Alfonso-Avila et al., 2017). They also are known to interact with each other and with other dietary elements and ingredients (Sanchez et al., 1994).

The objective of current study was to evaluate the effect of supplementing two new formulas of dietary buffer with a high buffering capacity contained Na or K on feed intake, rumen pH, nutrient digestion, blood metabolite, milk production, and feed efficiency of dairy cow. The buffering capacity of both buffers was known to exceed the buffering capacity of $NaHCO_3$. We hypothesized that increased buffering capacity of supplemental dietary buffer would improve performance and health indicators of dairy cows and that the effects would be dependent on the mineral cation of the buffer.

2. Materials and methods

The experiment was conducted at the dairy farm of the Department of Animal Science, University of Tehran (Karaj, Iran) from August, 2017 through October, 2017. Animals were cared for according to the guidelines of the Iranian Council of Animal Care (1995).

2.1. Animals, experimental design, and treatments

Nine multiparous mid-lactation Holstein dairy cows (594 ± 46 kg BW; mean ± SD) averaging 120 ± 28 days in milk (with a range of 87–154 days in milk) and producing 46.6 ± 3.4 kg/d of milk were used in a replicated 3 × 3 Latin square design with 21-d periods. Each period consisted of 14 d of adaptation followed by 7 d of data collection. During each 21-d period, cows were offered one of three total mixed rations that varied in dietary buffer. The three types of dietary buffer were 1) 11.2 g/kg of dietary dry matter (DM) sodium bicarbonate (SB; control) 2) 8.7 g/kg of dietary DM high buffering capacity formula contained Na (HBNa; Pishgam Damparvar Sepahan Co., Isfahan, Iran), and 3) 7.4 g/kg of dietary DM high buffering capacity formula contained K (HBK; Pishgam Damparvar Sepahan Co., Isfahan, Iran). The amount of administration was based on company recommendation. Diets were formulated to meet or exceed the Cornell Net Carbohydrate and Protein System (CNCPS, version 5.0; Fox et al., 2000) nutrient allowance for a lactating dairy cow weighing 680 kg and producing 46 kg/d (Table 1). All cows were housed in individual tie stalls with concrete floors that were cleaned regularly and had free access to water at all times. Feed was supplied twice daily at 0830 and 1630 h in amounts that allowed 5–10% refusals. Diets were manually mixed and weighed into each cow's feed trough, and refusals were manually removed daily.

2.2. Intake, digestibility and chemical analyses

The diet amounts offered and refused were measured daily for each cow during days 15 to 19 of each period and daily dry matter intake (DMI) for each cow was calculated. Representative samples of treatment diets (pooled by diet within period), and individual refusals

Table 1
Ingredient nutrient content of treatment diets[a].

Item	Treatments		
	SB	HBNa	HBK
Ingredients, g/kg of DM			
Corn silage	229	229	229
Alfalfa hay	146	146	146
Beet pulp	70	70	70
Corn grain, ground	185	185	185
Barley grain, ground	153	153	153
Soybean meal	112	112	112
Extruded soybean	26	26	26
Corn gluten meal	11	11	11
Fish meal	18	18	18
Fat supplement	21	21	21
Wheat bran	4.0	6.0	7.3
Buffer	11.2	8.7	7.4
Mgo	2.8	2.8	2.8
Trace mineral and Vitamin mix[b]	4.7	4.7	4.7
Calcium carbonate	4.2	4.2	4.2
Dicalcium phosphate	2.0	2.0	2.0
NaCl	1.4	1.4	1.4
Nutrient composition, g/kg of DM (otherwise stated)[c]			
DM, g/kg of as fed	503	494	497
Ash	71.9	68.3	72.8
CP	149	148	147
NDF	280	286	286
EE	43	42	41
NFC[d]	456	456	453
NEL, Mcal/kg of DM[e]	1.68	1.68	1.68

[a] Treatments were three types of dietary buffer: SB = sodium bicarbonate, HBNa = a high buffering capacity buffer contained Na, and HBK = a high buffering capacity buffer contained K.
[b] Contained 800 mg/kg of Fe, 3000 mg/kg of Cu, 10,000 mg/kg of Mn, 120 mg/kg of Co, 16,000 mg/kg of Zn, 80 mg/kg of Se, 150 mg/ kg of I, 2000 mg/kg monensin, 1300 kIU/kg of vitamin A, 360 kIU/kg of vitamin D, and 12 kIU/kg of vitamin E.
[c] DM = dry matter; CP = crude protein; NDF = neutral detergent fiber; EE = ether extract; NFC = non-fiber carbohydrates.
[d] NFC = nonfiber carbohydrate calculated by difference 100 − (% NDF + % CP + % Fat + % ash).
[e] Based on tabular values (CNCPS; Fox et al., 2000).

(pooled by cow within period) were taken during days 15 to 19 of each period. The DM concentration of diets and refusals samples was determined by drying at 60 °C in a forced-air oven for 72 h. All samples were ground using a Wiley mill through a 1-mm screen (Retsch GmbH 5657 HAAN, Germany) and analyzed for crude protein (CP) using the Kjeldahl method (Kjeltec 1030 Auto Analyzer, Tecator, Höganäs, Sweden; AOAC, 2002, method 955.04), ether extract (EE; AOAC, 2002, method 920.39), ash and organic matter (OM) (AOAC, 2002; method 942.05), and neutral detergent fiber (NDF) using heat stable α-amylase (100 μL/0.5 g of sample, Van Soest et al., 1991). Non-fiber carbohydrates (NFC) was calculated as 100 − [CP + NDF + EE + ash]. Refusals from individual cows were used for calculation of nutrient intake.

Buffering capacity for the three buffers was measured based on titration method (Jasaitis et al., 1987) with 2 N HCl and the buffering capacity of each buffer was expressed as percentage of a standard $NaHCO_3$. The concentrations of K, Na, and S in three buffers and relative diets were determined by inductively coupled plasma optical emission spectrometry (ICP-OES; Varian 735-ES, Palo Alto, CA, USA). Cl concentration was determined by potentiometric titration and ICP-OES.

Six fecal grab samples were collected from each cow at 0030, 0830, and 1630 of days 17 and 18 to represent the 24-h feeding cycle. Fecal samples were composited by period and analyzed for nutrient composition. Apparent total-tract digestibilities of DM, OM, EE, NDF, and NFC were determined using acid-insoluble ash (AIA) as an internal marker to calculate apparent digestibility using the 2 N HCl procedure of Van

Keulen and Young (1977).

2.3. Chewing activity and rectal temperature

On days 15 and 16 of each period, chewing activity was monitored visually for all cows over a 24-h period except for milking time. Eating and ruminating activities were noted by an observer at 5-min intervals, and each activity was assumed to persist for the entire 5-min interval. On day 15 of each period, rectal temperature was measured at about 1500 h using a clinical veterinary thermometer inserted in the rectum.

2.4. Rumen and blood sampling, and analyses

On day 21 of each period, 4 h after the morning feeding, rumen fluid (approximately 3 mL) from the ventral sac was sampled via rumenocentesis (Nordlund and Garrett, 1994). The pH of the ruminal fluid was immediately determined using a portable digital pH meter (HI 8318; Hanna Instruments, Cluj-Napoca, Romania).

On day 15 of each period, blood samples (7 mL) were collected before the morning feeding via the coccygeal vein using an evacuated tube without anticoagulant. Blood samples were placed on ice immediately after collection and centrifuged at $3000 \times g$ for 15 min. Serum samples were separated and stored in plastic tubes frozen at $-10\,°C$ until analysis. The concentrations of serum glucose, cholesterol, blood urea-N (BUN), high density lipoprotein (HDL) cholesterol, triglyceride (TG), total protein, albumin, aspartate aminotransferase (AST), alkaline phosphatase (ALP), Ca, Mg, and Cl were measured by an autoanalyzer (Abbott Alycon 300, USA) using commercial kits (Pars Azmoon Co., Tehran, Iran) according to the manufacturer's instructions. The analyzer was calibrated and controls assayed daily according to the manufacturer's instructions to ensure acceptable assay performance. Total antioxidant capacity (TAC) was determined by commercial colorimetric kits (Randox Laboratories Ltd., Ardmore, UK) using the same autoanalyzer. The concentration of serum malondialdehyde (MDA) was determined by thiobarbituric acid reacting substances method, in which the absorbance of a colored complex that is formed from the reaction of MDA with 2-thiobarbituric acid in acid environment is measured at 532 nm (Wullepit et al., 2012). Globulin concentrations were calculated by subtracting albumin concentrations from total protein. The Na and K concentrations were determined by flame photometry, using a Corning 410 Flame Photometer (Ciba Corning Diagnostics Scientific Instruments, Essex, England).

2.5. Milk yield and components, and body weight

Cows were milked three times daily at 0000, 0800, and 1600 h in a herringbone milking parlor. Milk yield was recorded during day 15 to 19 of the study and milk samples were collected from each milking during day 15 to 17 of the study. Milk samples were preserved with potassium dichromate and stored at $4\,°C$ until further analysis for fat, true protein, lactose, and solid not fat content (Fossomatic5000, Foss Electric, Hillerød, Denmark). Milk yield was corrected for milk components as 3.5% fat corrected milk (FCM = 0.432 milk yield + 13.23 fat yield; Gaines, 1928), energy corrected milk (ECM = 0.3246 milk yield + 12.96 fat yield + 7.04 protein yield; Jenkins et al., 1998), and solid corrected milk (SCM = 12.3 fat yield + 6.56 solid not fat yield - 0.0752 milk yield; Tyrrell and Reid, 1965). Cows were weighed at the beginning (d 1) and the end (d 20) of each period.

2.6. Statistical analyses

Data were analyzed using the mixed model procedure of SAS (Proc Mixed; SAS Institute, 2002) to account for effects of square, cow within square, and treatment. The treatment, square, and period were considered as fixed effects; cow within square was considered a random effect. Estimation method was REML and the degrees of freedom

Table 2
Buffering capacity of buffers and mineral composition of buffers[a] and diets.

	SB	HBNa	HBK
Buffering capacity % of pure NAHCO$_3$	101.6	149.7	137.4
Composition			
Buffers			
Na, g/kg	272	305	249
K, g/kg	0	0	58
Cl, g/kg	1	8	9
S, g/kg	1	0	0
Diets			
Na, g/kg	4.7	4.3	3.5
K, g/kg	12.5	12.5	12.9
Cl, g/kg	11	1.1	1.1
S, g/kg	2.0	2.0	2.0
DCAD[b]	369	350	327

[a] Treatments were three types of dietary buffer: SB = sodium bicarbonate, HBNa = a high buffering capacity buffer contained Na, and HBK = a high buffering capacity buffer contained K.
[b] Dietary cation-anion difference.

method was Kenward-Rogers. The differences among the treatments were evaluated using a multiple comparison test following the Tukey-Kramer method. Statistical significance of treatment was declared at $P \le 0.05$, and tendencies were discussed at $0.05 \le P \le 0.10$.

3. Results

3.1. Diets and buffers

The analyzed chemical composition was similar among the three diets (Table 1). On average, the diets contained 150 g/kg CP, 455 g/kg NFC, and 1.68 Mcal/Kg NEL on a dry matter basis.

The buffering capacities of HBNa buffer (149.7%) and HBK buffer (137.4%) were greater than SB buffer (101.4%; Table 2). The concentrations of Na was highest for HBNa buffer (305 g/kg), medium for SB buffer (272 g/kg) and lowest for HBK buffer (249 g/kg). Buffer HBK contained 58 g/kg K, whereas buffers SB and HBNa contained 0 g/kg K. Buffers SB, HBNa, and HBK contained 1, 8, and 9 g/kg Cl, respectively. Among the three buffers, SB only contained a 1 g/kg S. The concentration of Na was 4.7, 4.3, and 3.5 g/kg for SB, HBNa, and HBK diets, respectively. While SB and HBNa diets contained 12.5 g/kg K, this value for HBK diets was 12.9 g/kg. All the three diets contained a similar amounts of Cl and S, and finally, the amount of DCAD was 369, 351 and 327 for SB, HBNa and HBK diets, respectively.

3.2. DMI, rumen pH, and nutrient digestibility

Intake of DM tended ($P = 0.06$) to be lower with HBK (20.6 kg/d) than SB (21.0 kg/d) and HBNa (21.2 kg/d; Table 3). Rumen pH of cows as well as digestibility of DM, OM, NDF, CP, EE, and NFC were not affected by treatments and in average rumen pH and DM digestibility in the total digestive tract were 5.88 and 79.4%, respectively.

3.3. Chewing activity, rectal temperature, and blood metabolites

Supplementation of different buffers did not affect daily eating, rumination, or chewing, or any of these measures normalized to DMI (Table 4). Cows fed HBNa and HBK diets tended ($P = 0.06$) to have a greater rectal temperature than those fed SB.

Cows fed HBK tended ($P = 0.08$) to have greater blood glucose concentration than other treatments, but concentrations of cholesterol, HDL, and triglyceride were not affected by treatments (Table 5). Concentrations of BUN, total protein, albumin, and globulin, activities of AST and ALP, and concentrations of MDA and TAC in serum also were similar among treatments. Concentration of Ca was greater for HBK

Table 3

Chewing activity for dairy cows fed diets differing in buffer supplementation (n = 9 cows).

Item	Treatments[a]			SEM	P-value
	SB	HBNa	HBK		Treat
Intake kg/d	21.0	21.2	20.6	0.73	0.06
Rumen pH	5.96	5.85	5.85	0.13	0.72
Apparent digestibility[b], %					
DM	79.9	79.0	79.4	1.69	0.86
OM	81.4	80.7	81.0	1.66	0.89
NDF	68.8	66.0	67.2	2.53	0.63
CP	88.3	88.1	88.7	1.10	0.87
EE	78.4	77.0	78.0	1.77	0.71
NFC	90.6	89.5	89.3	1.12	0.33

[a] Treatments were three types of dietary buffer: SB = sodium bicarbonate, HBNa = a high buffering capacity buffer contained Na, and HBK = a high buffering capacity buffer contained K.

[b] DM = dry matter; OM = organic matter; NDF = neutral detergent fiber; CP = crude protein; EE = ether extract; NFC = non-fiber carbohydrates.

Table 4

Chewing activity for dairy cows fed diets differing in buffer supplementation (n = 9 cows).

Item	Treatments[a]			SEM	P-value
	SB	HBNa	HBK		Treat
Eating					
Min/d	231	261	244	16.9	0.14
Min/kg of DM	11.0	12.4	11.9	0.83	0.20
Ruminating					
Min/d	434	419	439	27.2	0.67
Min/kg of DM	31.8	32.5	33.5	1.69	0.44
Total chewing					
Min/d	664	680	683	24.3	0.72
Min/kg of DM	31.8	32.5	33.5	1.69	0.44
Rectal temperature, °C	38.0	38.3	38.4	0.14	0.06

[a] Treatments were three types of dietary buffer: SB = sodium bicarbonate, HBNa = a high buffering capacity buffer contained Na, and HBK = a high buffering capacity buffer contained K.

(10.4 mg/dL) than for SB (9.74 mg/dL) or HBNa (9.93 mg/dL; $P = 0.01$). Additionally, HBK and HBNa diets tended to induce greater concentration of Cl in serum than the SB diet, whereas concentrations of Na and K were not affected treatments.

3.4. Milk yield and composition, feed efficiency, and body weight change

Yields of milk, 3.5% FCM, ECM, and protein were not affected by treatments, but SCM yield and fat yield tended to be greater in HBK than in HBNa and SB ($P = 0.7$). Milk fat concentration also was greater in cows fed HBK (32.5 g/kg) than other treatments (29.5 and 29.6 for SB and HBNa, respectively; $P = 0.04$), and the percentage of protein, lactose, and SNF was greater in cows fed HBK than those fed SB ($P = 0.02$; Table 6).

The efficiency of milk yield was greater ($P = 0.01$) in cows fed HBK (1.86) than in cows fed SB (1.80), whereas HBNa (1.82) did not improve milk yield efficiency (Table 6). The efficiency of SCM yield also was greater (1.64 vs. 1.51 and 1.15; $P = 0.02$) and efficiency of ECM yield tended ($P = 0.07$) to be greater in HBK than SB and HBNa. Body weight and body weight change were not affected by treatments.

4. Discussion

Achieving the dietary buffer requirement of dairy cows is a complex task. Complexity results from the interactions of dietary acid load and electrolyte balance that affect rumen and blood pH (Erdman, 1988;

Table 5

Blood metabolites in dairy cows fed diets differing in in buffer supplementation (n = 9 cows).

Item[a]	Treatments[b]			SEM	P-value
	SB	HBNa	HBK		Treat
Glucose, mg/dL	58.4	58.8	64.7	2.09	0.08
Cholesterol, mg/dL	242	240	243	18.3	0.93
HDL, mg/dL	47.9	47.9	46.6	3.50	0.91
Triglyceride, mg/dL	14.7	13.8	15.7	0.80	0.10
BUN, mg/dL	17.8	18.0	17.9	1.00	0.97
Total protein, g/dL	8.82	9.06	9.00	0.17	0.18
Albumin, g/dL	3.70	3.72	3.72	0.11	0.92
Globulin, g/dL	5.12	5.33	5.24	0.20	0.41
AST, U/L	71.1	74.3	66.7	5.24	0.58
ALP, U/L	109	122	115	7.92	0.34
TAC, mmol/L	0.52	0.50	0.53	0.029	0.75
MDA, μmol/L	1.01	1.06	1.11	0.09	0.73
Ca, mg/dL	9.74[b]	9.93[b]	10.37[a]	0.27	0.01
Mg, mg/dL	2.12	2.03	2.03	0.06	0.24
Na, mmol/L	166	162	167	3.90	0.64
K, mmol/L	4.99	4.68	4.64	0.174	0.30
Cl, mmol/L	101	104	104	1.20	0.09

[a,b] Least squares means within a row with different superscripts differ significantly (P < 0.05).

[a] HDL = high density lipoprotein; BUN = plasma urea N; AST = aspartate aminotransferase; ALP = alkaline phosphatase; TAC = total antioxidant capacity; MDA = malondialdehyde.

[b] Treatments were three types of dietary buffer: SB = sodium bicarbonate, HBNa = a high buffering capacity buffer contained Na, and HBK = a high buffering capacity buffer contained K.

Table 6

Milk production and composition, feed efficiency, and body weight change in dairy cows fed diets differing in buffer supplementation (n = 9 cows).

Item	Treatments[a]			SEM	P-value
	SB	HBNa	HBK		Treat
Yield, kg/d					
Milk	38.4	38.0	38.0	1.74	0.74
3.5% FCM[b]	31.8	31.3	32.9	1.62	0.21
ECM[c]	35.7	35.2	37.1	1.71	0.16
SCM[d]	31.7	31.5	33.7	1.47	0.07
Fat	1.14	1.13	1.24	0.08	0.10
Protein	1.18	1.18	1.22	0.05	0.14
Composition, g/kg					
Fat	29.5[b]	29.6[b]	32.5[a]	1.6	0.04
Protein	30.6[b]	31.0[ab]	32.2[a]	0.6	0.02
Lactose	44.4[b]	45.1[ab]	46.8[a]	0.9	0.02
SNF	81.0[b]	82.2[ab]	85.3[a]	1.5	0.02
Fat:protein	9.6	9.6	10.1	0.5	0.20
Feed efficiency					
Milk yield/DMI	1.80[b]	1.82[ab]	1.86[a]	0.10	0.01
ECM/DMI	1.51	1.49	1.60	0.07	0.07
SCM/DMI	1.51[b]	1.51[b]	1.64[a]	0.07	0.02
BW, kg	602	600	607	17.0	0.36
BW changes, kg/period	−4.16	−2.83	4.00	4.98	0.48

[a,b] Least squares means within a row with different superscripts differ significantly (P < 0.05).

[a] Treatments were three types of dietary buffer: SB = sodium bicarbonate, HBNa = a high buffering capacity buffer contained Na, and HBK = a high buffering capacity buffer contained K.

[b] 3.5% FCM (fat corrected milk) = 0.432 milk yield + 13.23 fat yield (Gaines, 1928).

[c] ECM (energy corrected milk) = 0.3246 milk yield + 12.96 fat yield + 7.04 protein yield (Jenkins et al., 1998).

[d] SCM (solid corrected milk) = 12.3 fat yield + 6.56 solid not fat yield - 0.0752 milk yield (Tyrrell and Reid, 1965).

Sanchez et al., 1994; Iwaniuk and Erdman, 2015). Therefore, assessing the dietary buffer requirement requires consideration of both acid neutralization and mineral balance, which is the goal of supplemental buffer formulation. The study tested increasing buffering capacity by adding a Na containing and a K containing buffer formula. In summary, performance was most improved by a supplemental K contained formula.

Our results indicated that increasing buffering capacity by adding HBNa did not improve nutrient intake, digestion, chewing behavior, rumen pH, blood metabolism, or milk production. Our results are not consistent with previous studies that have demonstrated improved rumen fermentation (Wu et al., 2015). Our observations might be explained by the inclusion of approximately 1% $NaHCO_3$ in the control diet. In a review of 27 studies, Hu and Murphy (2005) demonstrated that a high level of $NaHCO_3$ supplementation induced similar responses in intake, digestion and milk production of dairy cows when compared with a moderate level of $NaHCO_3$ supplementation. They proposed that addition of $NaHCO_3$ at 0.7–1% of dietary DM was optimal for early- and mid-lactation cows fed corn silage based diets. Responses of rumen pH to varied levels of supplemental $NaHCO_3$ also indicated that increasing buffering capacity did not additively increase rumen pH (Hu and Murphy, 2005). Erdman (1988) indicated that buffers containing a high amount of Na are extremely hygroscopic and prone to caking. We also observed this characteristic in HBNa buffer, so it is possible that performance of the buffer in the present study was limited by physical form.

The null effect on rumen pH and apparent nutrient digestibility between HBK and SB could also be explained by condition of control diet which contained about 1% NaHCO3. Previous studies have indicated that buffers containing Na or K when fed at levels near 1–2 % of dietary DM have similar effects on rumen pH or nutrient digestion (West et al., 1987; Alfonso-Avila et al., 2017). In our study, we also found that the three treatments similarly affected rumen pH and nutrient digestion. In contrast to previously reported work (West et al., 1986; Iwaniuk et al., 2015; Alfonso-Avila et al., 2017), the HBK diet tended to decrease DMI. Treatment HBK attended to induce greater concentration of glucose in serum which has been proposed to contribute to satiety (Allen, 2000). To our knowledge, no mechanism relating blood glucose to dietary buffer has been previously reported.

Ca was the only mineral in the blood that was elevated by feeding HBK. Previous studies have reported that greater concentration of Ca in milk is induced by supplementation of K_2CO_3 rather than Na_2CO_3 but no mechanism was proposed (Alfonso-Avila et al., 2017). However, Ca concentration in plasma may be inversely correlated with metabolic acidosis as Ca is less deposited to bone (Sanchez et al., 1994). The effect of BK on Ca concentration in serum observed in our study could therefore be attributed to improved electrolyte balance in blood.

Greater percentage of fat, protein, and lactose in HBK was associated with greater SCM yield. The effect of K on rumen biohydrogenation products has been recently documented to improve milk fat concentration (Alfonso-Avila et al., 2017). Improvement of milk fat percentage also has been prominent in previous studies comparing buffers with K or Na mineral cations (West et al., 1986; Staples et al., 1988; Iwaniuk et al., 2015). Supplementation of buffer with K cation has increased milk fat in conditions of similar or lower rumen pH than reported in our study (West et al., 1986; Alfonso-Avila et al., 2017; Staples et al., 1988) because it modulates the intermediate factors relating milk fat depression and rumen pH (i. g. trans-10, cis-12 CLA; Jenkins et al., 2014). It is suggested that K promotes the predominant trans-11 biohydrogenation pathway rather than the trans-10 pathway in the rumen (Ma et al., 2017). Effects of such K-based buffers on protein percentage, however, have been inconsistent (West et al., 1986; Mooney and Allen, 2007; Staples et al., 1988; Alfonso-Avila et al., 2017).

The cellular mechanisms involved in the transport of milk constituents out of and across the mammary epithelium is composed of 4 transcellular routes and one paracellular route (Shennan and Peaker, 2000). These mechanisms require that a constant concentration of gradients in body fluids be maintained to support transport of substrates or metabolites in and out of cells, as well as regulation of osmotic pressure (McDowell, 1992). A previous study that reported greater milk protein percentage with K administration used a multi-element buffer (Staples et al., 1988), which is similar to the HBK used in our current study. Because numerous minerals affect osmotic balance (Sanchez et al., 1994), it could be postulated that the effects of HBK in the current study might be due to the interaction among the minerals contained. However, more work is need to elucidate the mechanism of action.

The improvement in feed efficiency induced by HBK was one important outcome of the current study. Because nutrient digestibility, body weight and body weight change were comparable among the three treatments, we assume that the change in feed efficiency is the result of improved nutritional status and nutrients metabolism by meeting the real requirement for K. Erdman et al., (2011) reported a similar effect of dietary K level on feed efficiency and indicated that, compared with other species, the ruminant requirement for K is greater because of rumen bacterial metabolism. The K requirement may be greater in stressed animals such as high milk producing cows fed a high concentrate diet (Li et al., 2012; Alfonso-Avila et al., 2017; Nasrollahi et al., 2017b), which was a condition of the present study. The requirement for K in high milk producing dairy cows perhaps should continue to be evaluated in relation to dietary acid load and electrolyte balance. Future study examining rumen microbiology, and internal metabolism are warranted to disclose the molecular background of the events as well as quantitative calculation of the requirement of recent generation of dairy cows.

Finally, it should be noted that the number of animals used in the current study, as well as the use of a short-term Latin square design (21 days), may affect the comparability of the final results with other studies and conditions of practice. Therefore, additional research with more animals in a longer term design is warranted to validate the results of this study.

5. Conclusion

Supplementation of HBNa to the diet of mid-lactation dairy cow was along with similar response on feed intake, nutrient digestion, feeding behavior, blood metabolites, milk production and feed efficiency when compared with SB. Supplementation of HBK, however, increased concentration of Ca in serum and tended to increase feed intake and blood glucose. Supplementation of HBK also improved milk composition and feed efficiency without affecting body weight change and nutrient digestibility. Under conditions of our study, buffer HBK with 6% K most improved milk composition and feed efficiency.

Conflict of interest statement

None of the authors had a personal or financial conflict of interest.

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