Intraruminal Infusion of Propionate Alters Feeding Behavior and Decreases Energy Intake of Lactating Dairy Cows[1,2]

Masahito Oba[3] and Michael S. Allen[4]

Department of Animal Science, Michigan State University, East Lansing, MI

ABSTRACT The dose-response effects of intraruminal infusion of propionate on feeding behavior of lactating dairy cows were evaluated with eight ruminally cannulated Holstein cows past peak lactation. Treatments were mixtures of propionic acid and acetic acid containing propionic acid at eight different concentrations in Experiment 1, and mixtures of sodium propionate and sodium acetate containing sodium propionate at 4 different concentrations in Experiment 2. Experimental designs were an 8 × 8 and duplicated 4 × 4 Latin squares, respectively, for Experiments 1 and 2. Treatment solutions were infused into the rumen continuously for 14 h at a rate of 16.7 and 25 mmol/min, respectively, for Experiments 1 and 2. Infusion started 2 h before feeding and ended 12 h after feeding; feeding behavior was monitored for 12 h after feeding using a computerized data acquisition system. Total metabolizable energy (ME) intake was calculated by adding the energy of infusates to dietary energy intake. In both experiments, as the proportion of propionate of the infusates increased, total ME intake and dry matter intake decreased linearly. As infusion of propionate increased, meal size tended ($P < 0.09$) to decrease linearly and intermeal interval tended ($P < 0.07$) to increase linearly in Experiment 1; meal size decreased linearly and number of meal bouts tended ($P < 0.08$) to decrease linearly in Experiment 2. These observations indicate that the reduction in dietary energy intake from propionate infusion was greater than the energy supplied from infusates, and that propionate plays an important role in feed intake regulation by affecting both satiety and hunger. J. Nutr. 133: 1094–1099, 2003.

KEY WORDS: • *propionate infusion* • *feed intake* • *feeding behavior* • *dairy cow*

Maximizing energy intake is an important goal for nutritional management of high producing dairy cows. Although feeding more fermentable grains in diets increases their energy density, excess fermentation in the rumen sometimes decreases dry matter intake (DMI)[5] and does not necessarily increase energy intake in lactating cows (1). However, greater ruminal fermentation is more desirable to increase microbial protein production as well as energy intake unless DMI is decreased. Therefore, it is important to understand mechanisms that regulate voluntary feed intake when cows are fed highly fermentable diets.

Greater ruminal fermentation increases the production of fermentation acids with a greater proportion of propionate. Choi and Allen (2) showed that propionate has greater hypophagic effects in lactating dairy cows compared with acetate. Hypophagic effects of propionate have been documented extensively for ruminants (3–9). However, some experiments in the literature reported that propionate infusion did not decrease feed intake (10–13). The inconsistent hypophagic effects of propionate might be explained by a threshold response of propionate for feed intake regulation. Infusion of propionate might not affect DMI and feeding behavior unless propionate flux exceeds a threshold. This concept agrees with observations that feeding more fermentable grains does not always decrease DMI in lactating dairy cows (1).

The dose-response effects of propionate on feed intake were investigated previously for lactating dairy cows (10) and sheep (5). However, it is difficult to interpret those results because propionate was infused for only 3 h; thus, the effects of propionate were evaluated essentially for meal size only. Hypophagic effects of propionate should be evaluated by monitoring feeding behavior for a longer period because cows are able to compensate for smaller meal size by increasing meal frequency (2). In addition, the majority of previous experiments that studied hypophagic effects of propionate by infusion focused on DMI rather than energy intake, although animals are supplied energy from infused propionate and maximization of energy intake is a primary concern for practical nutritional management.

Little evidence exists in the literature regarding the effects of propionate on energy intake and feeding behavior. The objectives of this experiment were to evaluate dose-response effects of intraruminal infusion of propionate on feeding behavior and energy intake in lactating dairy cows and to deter-

[1] Presented in part at the American Dairy Science Association annual meeting, July 2001, Indianapolis, IN [Oba, M. & Allen, M. S. (2001) Dose-response effects of intra-ruminal infusion of propionate on feeding behavior of lactating dairy cows. J. Dairy Sci. 84(Suppl. 1): 122 (abs.)].

[2] Supported by the Michigan Agricultural Experiment Station, East Lansing, Michigan.

[3] Present address: Department of Animal and Avian Sciences, University of Maryland, College Park, MD.

[4] To whom correspondence should be addressed. E-mail: allenm@msu.edu.

[5] Abbreviations used: DMI, dry matter intake; ME, metabolizable energy; VFA, volatile fatty acids.

Manuscript received 6 August 2002. Initial review completed 20 October 2002. Revision accepted 23 December 2002.

mine whether a threshold exists for the effects of propionate infusion on feed intake. We hypothesized that propionate infusion decreases energy intake by reducing meal size and possibly meal frequency, and hypophagic effects of propionate increase with infusion rate.

MATERIALS AND METHODS

Experimental procedures were approved by the All University Committee on Animal Use and Care at Michigan State University.

Experiment 1. Multiparous Holstein cows ($n = 8$; 113 ± 26 d in milk; mean ± SD) cannulated ruminally for previous experiments were selected from the Michigan State University Dairy Cattle Teaching and Research Center. Treatments were continuous intraruminal infusion of mixtures of propionic acid and acetic acid at eight different ratios. Treatment solutions were prepared by diluting 16.8 mol of volatile fatty acids (VFA) to 18 L with deionized water. Concentrations of propionic acid were 0, 0.14, 0.29, 0.43, 0.57, 0.71, 0.86 and 1.00 as a fraction of total VFA infused. Acetic acid was added to infusates to isolate specific effects of propionate relative to acetate. Concentrations of total VFA were 0.93 mol/L across the treatments, and 15 L of each solution was infused over 14 h. Infusion rate was 17.9 mL/min, which is equivalent to the infusion of 16.7 mmol VFA/min. This rate of infusion was approximately one third the estimated rate of VFA production for lactating dairy cows consuming 10 kg/d of ruminally fermented organic matter (14). The solutions were infused using 4-channel peristaltic pumps (#78016–30, Cole-Parmer Instrument, Vernon Hills, IL) and Tygon tubing (7.5 m × 1.6 mm i.d.; Fisher Scientific Co., Pittsburgh, PA). Infusion started 2 h before feeding so that treatments could influence feeding behavior from the first meal immediately after feeding. Treatment periods were 2 d, with 14 h of infusion followed by 34 h of recovery.

The experimental diet contained dry cracked corn (mean particle size of 3.6 mm), corn silage, alfalfa silage, a premix of protein supplements (soybean meal, distillers grains, and blood meal) and a premix of minerals and vitamins (**Table 1**). Dietary neutral detergent fiber, crude protein and starch concentrations were 290, 159 and 308 g/kg, respectively. Dry cracked corn was the major source of starch to minimize propionate production from the basal diet. The diet adaptation period was 14 d, and the final 3 d were used for data collection for DMI and milk yield to characterize the cows used in this experiment. Body weight was determined on the last day of the diet adaptation period. Means for body weight, DMI and milk yield were 623 kg, 25.4 kg/d and 36.4 kg/d, respectively. After 14 d of diet adaptation, cows were assigned to an 8 × 8 Latin square balanced for carry-over effects of infusion treatments.

TABLE 1

Ingredients and nutrient composition of experimental diet (g/kg of dietary DM except for DM; DM expressed as g/kg of diet as fed)

Diet Ingredients	
Corn silage	270
Alfalfa silage	254
Dry cracked corn	259
Whole linted cottonseed	68
Protein mix[1]	99
Vitamin & mineral mix[2]	50
Nutrient Composition	
DM	495
OM	930
Starch	308
Neutral Detergent Fiber	290
Acid Detergent Fiber	208
Crude Protein	159
Ether extract	38
Forage Neutral Detergent Fiber	212
Metabolizable energy, *MJ/kg*[3]	11.4

[1] Protein mix contained 750 g/kg soybean meal, 200 g/kg distillers grain, and 50 g/kg blood meal.

[2] Vitamin & mineral mix contained 5.3 g/100 g Ca, 4.4 g/100 g P, 1.6 g/100 g Mg, 0.3 g/100 g K, 3.2 g/100 g Na, 5.0 g/100 g Cl, 0.3 g/100 g S, 8 μg/g Co, 243 μg/g Cu, 1501 μg/g Fe, 14 μg/g I, 1065 μg/g Mn, 6 μg/g Se, 1018 μg/g Zn, 109,000 IU/kg vitamin A, 27,500 IU/kg vitamin D, and 445 IU/kg vitamin E.

[3] Metabolizable energy was calculated from book values according to NRC (16).

Throughout the experiment, cows were housed in tie-stalls, and fed once daily (1030 h) at 110% of expected daily intake. Cows were not allowed access to feed from 0830 to 1030 h. The amount of feed offered and orts were weighed for each cow daily. On every infusion day, samples of all dietary ingredients (0.5 kg) were collected, and treatment solutions were infused from 0830 to 2230 h. Cows were milked twice daily in the milking parlor except for the evening milking on infusion day, for which cows were milked in their stalls. Feeding behavior was monitored from 1030 to 2230 h on every infusion day by a computerized data acquisition system (15), and feed was always available when feeding behavior was monitored. Data on chewing activities, feed disappearance and water consumption were recorded for each cow every 5 s, and meal bouts, interval between meals, meal size, eating time, ruminating time and total chewing time were calculated. Independent meal bouts were defined using the following criteria: minimum eating chews of 0.45/s over a 180-s period and 0.3/s over a 30-s period, minimum feed disappearance of 50 g, minimum interval of 7.5 min from a previous meal (15). Independent drinking bouts were defined as minimum interval of 4.0 min from a previous bout (15). Total metabolizable energy (ME) intake was calculated by adding the ME from infusates to the ME from the diet. The experimental diet was assumed to contain 11.4 MJ/kg of ME on the basis of book values from the NRC (16). Intake of ME was evaluated instead of intake of net energy for lactation because the efficiency of energy conversion from ME to net energy for lactation for infused acetate and propionate is not known and it could be different depending on milk fat concentration. In addition, the effect of propionate on energy intake was our primary concern, and evaluation of ME intake was appropriate to accomplish the objectives of this experiment. Acetate and propionate were assumed to contain 0.2094 and 0.3672 Mcal/mol of ME, respectively (8). Infusates were weighed before and after infusion, and actual amount of solutions infused into the rumen was calculated. The ME from infusates was calculated by multiplying ME concentration of infusates by the amount infused into the rumen for 12 h.

Diet ingredients were dried in a 55°C forced-air oven for 72 h and analyzed for DM concentration. All samples were ground with a Wiley mill (1-mm screen; Authur H. Thomas, Philadelphia, PA). Samples were analyzed for ash, neutral detergent fiber, acid detergent fiber, crude protein and starch. Ash concentration was determined after 5 h of oxidation at 500°C in a muffle furnace. The neutral detergent fiber and acid detergent fiber concentrations were determined [(17); method A for neutral detergent fiber]. Crude protein was analyzed according to Hach et al. (18). Starch was measured by an enzymatic method (19) after samples were gelatinized with sodium hydroxide; glucose concentration was measured using a commercial kit (Glucose kit #510; Sigma Chemical, St. Louis, MO). Concentrations of all nutrients except DM were expressed as percentages of DM determined from drying at 105°C in a forced-air oven. Corn grain was dry-sieved through 8 sieves (sieve apertures: 4750, 2360, 1180, 600, 300, 150, 75 μm and bottom pan), using a sieve shaker (Model RX-86, W.S. Tyler, Gastonia, NC) for ~20 min until the bottom pan weight was constant; then, mean particle size of corn grain was calculated (20).

All data for Experiment 1 were analyzed using the fit model procedure of JMP (version 4.0, SAS Institute, Cary NC) according to the following model:

$$Y_{ijkl} = \mu + C_i + P_j + L_k + Q_k + e_{ijkl}$$

in which Y_{ijkl} is a dependent variable, μ is overall mean, C_i is random effect of cow ($i = 1$ to 8), P_j is fixed effect of period ($j = 1$ to 8), L_k is linear effect of treatment, Q_k is quadratic effect of treatment and e_{ijkl} is the residual error. The actual amount of solution infused into the rumen was not affected by treatments and was not included in the

statistical model. Treatment effects were declared significant at $P < 0.05$, and tendency for treatment effects was declared at $P < 0.10$.

Experiment 2. The cows used for Experiment 1 were used for Experiment 2 at a later stage of lactation (159 ± 26 d in milk; mean ± SD). Cows were fed the same diet described for Experiment 1, and were assigned to duplicated 4 × 4 Latin squares balanced for carryover effects. Treatments were continuous intraruminal infusion of mixtures of sodium propionate and sodium acetate at four different ratios. The infusion rate of VFA for Experiment 2 was higher than that for Experiment 1 (25.0 vs. 16.7 mmol/min). Because of the higher infusion rate, acetate and propionate were infused as sodium salts rather than acids for Experiment 2 to avoid risk of ruminal acidosis. Treatment solutions were prepared by diluting 25.2 mol of VFA salt to 18 L with deionized water. Concentrations of propionate were 0, 0.33, 0.67 and 1.00 as a fraction of total VFA infused. Sodium acetate was added to infusates to isolate specific effects of propionate relative to acetate. Concentration of total VFA was 1.4 mol/L across the treatments, and 15 L of each solution was infused over 14 h. Infusion rate was 17.9 mL/min, which is equivalent to the infusion of 25.0 mmol VFA/min. This rate of infusion is approximately one half the estimated rate of VFA production for lactating dairy cows consuming 10 kg/d of ruminally fermented organic matter (14). Infusion protocol, methods for data and sample collection, methods for data and sample analysis were as described for Experiment 1.

All data for Experiment 2 were analyzed using the fit model procedure of JMP (version 4.0, SAS Institute, Cary, NC) according to the following model:

$$Y_{ijklm} = \mu + S_i + C(S)_{j(i)} + P_k + L_l + Q_l + Cov_{INF} + e_{ijklm}$$

in which Y_{ijklm} is a dependent variable, μ is the overall mean, S_i is the fixed effect of square ($i = 1$ to 2), $C(S)_{j(i)}$ is the random effect of cow nested in a square ($j = 1$ to 4), P_k is the fixed effect of period ($k = 1$ to 4), L_l is the linear effect of treatment, Q_l is the quadratic effect of treatment, Cov_{INF} is the effect of actual amount of solution infused into the rumen as a covariate and e_{ijklm} is the residual error. One pump was used for each square of 4 cows, and the random effect of cow was nested within squares that shared the same infusion pump. Interactions of square × treatment and period × treatment were originally evaluated, but they were removed from the statistical model because interactions were not significant for response variables of interest. The actual amount of solution infused into the rumen was included in the statistical model as a covariate because actual amount of infusates tended to differ by treatments (quadratic effect of treatments: $P < 0.09$). Linear and quadratic effects of treatments were evaluated. Treatment effects was declared significant at $P < 0.05$, and tendency for treatment effects was declared at $P < 0.10$.

RESULTS

Experiment 1. As infusion rate of propionate increased, DMI decreased linearly from 15.1 kg/12 h for 0% propionate to 13.2 kg/12 h for 100% propionate treatment ($P < 0.01$; **Table 2**). Intermeal interval tended to be longer ($P < 0.07$) and meal size tended to be smaller ($P < 0.09$) as the dose of propionate increased. Estimated total ME intake decreased linearly by infused propionate from 181.6 MJ/12 h for 0% propionate to 168.2 MJ/12 h for 100% propionate ($P < 0.05$). Eating time and ruminating time were not affected by treatment. Quadratic effects of treatment were not observed for any response variable.

Experiment 2. As the proportion of propionate in infusate increased, DMI decreased linearly from 15.0 kg/12 h for 0% propionate to 8.3 kg/12 h for 100% propionate treatment ($P < 0.001$; **Table 3**). Similarly, propionate decreased meal size linearly from 2.5 kg for 0% propionate to 1.5 kg for 100% propionate treatment ($P < 0.03$). The number of meal bouts over 12 h tended to decrease linearly ($P < 0.08$) with increasing propionate, although intermeal interval was not significantly affected by treatment. The estimated total ME intake decreased linearly by propionate dose from 186.2 MJ/12 h for 0% propionate to 121.8 MJ/12 h for 100% propionate ($P < 0.001$). Similarly, eating time decreased linearly, but rumi-

TABLE 2

Dose response effects of intra-ruminal infusion of propionate relative to acetate on feeding behavior of lactating dairy cows for experiment 1

	Propionate as a fraction of total VFA infused									*P* value	
	0	0.14	0.29	0.43	0.57	0.71	0.86	1.00	SEM	L[1]	Q[2]
Actual volume infused, *L*	14.7	14.6	14.8	14.8	14.5	14.7	14.8	14.8	0.2	0.45	0.79
Feeding behavior											
DMI, *kg/12 h*	15.1	14.9	14.3	15.2	13.5	13.0	13.6	13.2	0.6	<0.01	0.87
Meal bouts, */12 h*	8.3	6.9	6.9	8.1	7.6	7.2	7.5	7.1	0.5	0.58	0.81
Intermeal interval, *min*	65.1	76.6	83.6	71.6	73.1	74.7	79.1	85.5	5.3	0.07	0.91
Meal size, *kg DM*	1.9	2.3	2.2	2.0	1.8	2.0	1.9	1.9	0.1	0.09	0.76
Metabolizable energy intake											
Diet, *MJ/12 h*	171.1	169.5	162.3	172.4	153.6	147.7	154.0	150.2	7.1	<0.01	0.87
Infusion, *MJ/12 h*	10.5	11.3	12.6	13.8	14.6	15.9	17.2	18.0	0.1	<0.001	0.64
Total, *MJ/12 h*	181.6	180.7	174.9	186.2	168.2	163.6	171.1	168.2	7.1	0.05	0.87
Chewing time											
Eating, *min/12 h*	158	163	148	173	160	159	149	149	9.5	0.36	0.32
min/kg DMI	10.6	10.9	10.5	11.4	11.8	11.6	10.9	11.4	0.6	0.11	0.31
Ruminating, *min/12 h*	220	195	209	223	219	211	232	228	12	0.11	0.57
min/kg DMI	15.0	13.4	14.8	15.0	16.8	16.3	17.4	18.5	1.5	<0.01	0.44
Total, *min/12 h*	379	377	357	411	379	373	381	377	14	0.83	0.72
min/kg DMI	25.8	25.6	25.4	27.3	28.7	26.9	28.2	30.0	1.8	<0.01	0.70
Drinking behavior											
Water intake, *L/12 h*	49.9	48.9	48.4	46.9	51.3	47.6	48.3	48.2	2.1	0.61	0.87
Drinking bouts, */12 h*	11.4	11.1	10.5	9.6	10.0	10.3	11.3	9.3	0.6	0.06	0.47
Drinking interval, *min*	67.9	65.6	70.3	69.6	89.3	66.9	62.9	74.5	8.3	0.66	0.43
Drink size, *L/bout*	5.0	4.6	4.8	5.3	6.3	5.2	4.8	5.8	0.4	0.13	0.63

[1] Linear effect of treatments.
[2] Quadratic effect of treatments.

TABLE 3

Dose response effects of intra-ruminal infusion of propionate relative to acetate on feeding behavior of lactating dairy cows for experiment 2

	Propionate as a fraction of total VFA infused					*P* value	
	0	0.33	0.67	1.00	SEM	L[1]	Q[2]
Actual volume infused, *L*	15.2	15.0	14.9	15.1	0.2	0.58	0.09
Feeding behavior							
DMI, *kg/12 h*	15.0	13.3	11.5	8.3	0.7	<0.001	0.26
Meal bouts, */12 h*	7.4	7.4	6.0	6.1	0.6	0.08	0.92
Intermeal interval, *min*	75.4	76.3	87.7	90.1	9.6	0.21	0.91
Meal size, *kg DM*	2.5	2.0	2.1	1.5	0.3	0.03	0.71
Metabolizable energy intake							
Diet, *MJ/12 h*	170.3	151.0	131.0	93.7	7.9	<0.001	0.26
Infusion, *MJ/12 h*	15.9	20.1	23.0	28.0	0.3	<0.001	0.33
Total, *MJ/12 h*	186.2	171.5	154.0	121.8	7.9	<0.001	0.27
Chewing time							
Eating, *min/12 h*	178	161	153	110	10	<0.001	0.23
min/kg DMI	12.0	11.9	13.1	13.2	0.9	0.07	0.78
Ruminating, *min/12 h*	104	94	95	71	10	0.34	0.47
min/kg DMI	7.0	7.1	8.2	8.2	1.3	0.34	0.99
Total, *min/12 h*	282	255	248	181	17	<0.01	0.25
min/kg DMI	19.0	18.9	21.2	21.4	1.8	0.11	0.90
Drinking behavior							
Water intake, *L/12 h*	103.9	97.0	90.7	82.6	3.7	<0.001	0.87
Drinking bouts, */12 h*	14.6	13.6	11.8	11.6	0.8	<0.01	0.59
Drinking interval, *min*	47.8	50.3	60.9	57.1	2.9	<0.01	0.31
Drinking size, *L/bout*	8.2	8.0	8.5	7.7	0.5	0.68	0.52

[1] Linear effect of treatments.
[2] Quadratic effect of treatments.

nating time was not affected by treatment. Infusion of propionate decreased water intake linearly from 103.9 L/12 h for 0% propionate to 82.6 L/12 h for 100% propionate treatment. This reduction of water intake was due to less frequent water intake ($P < 0.01$) because water consumed per bout was not affected by treatment. Quadratic effect of treatment was not observed for any response variable.

DISCUSSION

Feed intake. In both experiments, infusion of propionate decreased DMI in a dose-dependent manner. Grovum (21) proposed that hypophagic effects of propionate can be mediated by increased osmotic pressure in the rumen. Some early studies reviewed by Grovum (21) used water infusion as a control for intraruminal VFA infusions. The hypophagic effects of propionate observed in those studies cannot be attributed to a specific effect of propionate because of a difference in osmotic pressure in the rumen, which was shown to affect feed intake during the first 10 min of a meal (22). In our study, acetic acid (Experiment 1) and sodium acetate (Experiment 2) were added to equalize the osmolarity and pH of infusates across treatments. In addition, infusion of sodium acetate is a better control for infusion of sodium propionate than that of NaCl for Experiment 2 because treatment effects on postabsorptive acid-base balance are also expected to be similar. Therefore, treatment effect in this study was attributed to the specific effect of propionate relative to acetate. Our results provide strong evidence for specific hypophagic effects of propionate, in agreement with previous studies. Infusion of propionate into the portal vein of sheep decreased feed intake to a greater extent compared with infusion of acetate or butyrate (3) and compared with infusion of acetate, mannitol or saline (5). Infusion of propionate into the mesenteric vein of steers reduced feed intake, whereas infusion of acetate did not (4).

An evaluation of feeding behavior is necessary to understand the regulation mechanism of DMI by propionate because DMI is a function of both meal size and intermeal interval, which are determined by satiety and hunger, respectively. Although the hypophagic effect of propionate has been investigated extensively, most experiments in the literature have monitored feed intake over very short periods ranging from 30 min to 3 h, and essentially investigated the effect of propionate on meal size only. In the present study, feeding behavior was monitored for 12 h, and effects of propionate on intermeal interval and meal frequency were evaluated as well as meal size. Infusion of propionate tended ($P < 0.09$) to decrease meal size and increase intermeal interval ($P < 0.07$) in Experiment 1, and decreased meal size and tended ($P < 0.08$) to decrease meal frequency in Experiment 2. Our observations indicated that propionate decreased feed intake by affecting both satiety and hunger. This is different than mechanisms related to osmotic effects; Choi and Allen (2) reported that intraruminal infusion of a hyperosmotic solution of NaCl decreased meal size but did not decrease DMI over a 12-h period because cows compensated for the smaller meal size by increasing meal frequency.

Grovum proposed that hypophagic effects of propionate are mediated by greater insulin secretion (21). Propionate stimulates insulin secretion in ruminants (23,24), and insulin can regulate feed intake for ruminants because insulin infusions were shown to decrease feed intake in sheep (11,25,26). However, Allen (1) argued that hypophagic effects of propionate are not mediated solely by insulin with the following evidence: *1*) hepatic denervation eliminated hypophagia caused by propionate infusion; *2*) propionate infusion decreased feed intake

without an increase in insulin for some experiments; and 3) hyperinsulinemic-euglycemic clamps do not decrease energy intake after accounting for glucose infused.

Energy intake. Decreased feed intake might be expected as propionate infusion increased relative to acetate because of the greater energy concentration of propionate compared with acetate. However, total ME intake also decreased linearly as propionate increased for both experiments. The reduction in ME intake from the diet exceeded that supplied from the infusate as the proportion of propionate increased. The magnitude of the reduction in total ME intake caused by propionate depends on the estimated value for dietary ME concentration; overestimation of dietary ME concentration can cause a greater reduction in estimated total ME intake. Therefore, total ME intake was also analyzed assuming 10% less dietary ME concentration (10.3 MJ/kg instead of 11.4 MJ/kg) to account for possible overestimation of dietary ME concentration. The assumption of 10% less ME concentration in the diet slightly reduced the significance of treatment effects for Experiment 1, resulting in a tendency for a treatment effect on total ME intake ($P = 0.06$; data not shown), but propionate infusion still resulted in a significant linear reduction in total ME intake for Experiment 2, which used a greater rate of VFA infusion ($P < 0.001$; data not shown). Previous reports have shown that propionate decreased DMI compared with an isocaloric infusion of a VFA mixture (6) or acetate (8) in lactating dairy cows. Wu et al. (9) reported lower DMI for cows infused with propionate into the duodenum compared with isocaloric infusion of glucose into the rumen. That is consistent with our results because glucose ferments to other VFA such as acetate and butyrate as well as to propionate. These studies, along with the present experiment, suggest that hypophagic effects of propionate cannot be explained simply by the additional energy supplied as propionate. Animals do not consume to meet their energy requirements per se but have specific mechanisms regulating satiety and hunger.

Increased ruminal fermentation has been related to reduced energy intake for lactating cows (1). McCarthy et al. (27) compared ground shelled corn and steam rolled barley in high grain diets containing >45% grain, and found that starch digestibility in the rumen was 77% for cows fed barley-based diets and 48.5% for cows fed corn-based diets. Propionate concentration in ruminal fluid was greater for barley-based diets than corn-based diets (31.0 vs. 26.4 mol/100 mol VFA). In their experiment, cows fed ground corn consumed 23.8 kg/d of DM, whereas cows fed steam rolled barley consumed 20.7 kg/d of DM. Although starch digestibility was lower for corn treatments, the amounts of DM and OM digested in the total tract appeared to be greater for cows fed ground corn because of greater DMI. Similarly, Overton et al. (28) fed ground shelled corn and steam rolled barley at five different ratios (100:0, 75:25, 50:50, 25:75 and 0:100, for ground shelled cornstarch:steam rolled barley starch) in low forage diets (45% dietary DM). They reported a linear increase in ruminal starch digestibility and propionate concentration in ruminal fluid, and a linear decrease in DMI as the ratio of steam rolled barley increased in the diet. In addition, the amounts of DM and OM apparently digested in the total tract decreased linearly as the fraction of steam rolled barley increased in the diets. Excess propionate production in the rumen might have limited energy intake as well as DMI when cows were fed very fermentable grains for both experiments.

Quadratic effects were not significant for any response variable in either experiment, providing no evidence for a threshold response to infused propionate for feeding behavior or energy intake in this study. Additionally, the breakpoint for response in DMI to treatments, estimated as $-b/2a$ for the regression equation of $ax^2 + bx + c$ (29), was not identified within the range of rate for propionate infusion for both experiments, indicating a linear hypophagic effect of propionate only. In agreement with our results, Anil et al. (10) and Farningham and Whyte (5) reported that infusion of propionate linearly decreased feed intake in a dose-dependent manner without a threshold. Leuvenink et al. (30) showed that propionate infusion into the mesenteric vein of mature sheep at a rate of 1 mmol/min did not decrease feed intake, but the infusion at a rate of 2 mmol/min significantly decreased feed intake. However, their data cannot be used to support a threshold response of propionate in feed intake because the effect of propionate was evaluated at only two levels of infusion and feed intake was decreased numerically for the lower dose although it did not differ from the control.

Water intake. Water intake was nearly twice as high in Experiment 2 compared with Experiment 1 (93.6 vs. 48.7 L/12 h). This difference is attributed to infusion of acids in Experiment 1 and sodium salts in Experiment 2. Murphy et al. (31) suggested that 1 g of sodium intake increases water intake by 0.05 L. Using this relationship, infusion of 483 g of sodium over 14 h in Experiment 2 would be expected to explain 24.2 L of the 44.9 L difference in water intake between Experiment 1 and Experiment 2. The infusion of sodium might have increased osmolarity of ruminal fluid, drawing water into the rumen from the blood and resulting in thirst and increasing water intake. In addition, greater water intake might be due to increased blood osmolarity and increased urine volume to excrete excess sodium. Although infusion treatments did not affect water intake in Experiment 1, water intake decreased linearly as propionate infusion increased in Experiment 2. Propionate might have had direct effects on thirst; however, this mechanism is not known and is highly speculative. This response is more likely because of the greater effect of propionate treatment on feed intake in Experiment 2 compared with Experiment 1 because infusates were isoosmotic within each experiment, and both feed intake and salt intake stimulate water intake in lactating dairy cows (31).

In conclusion, intraruminal infusion of propionate decreased DMI and ME intake in a dose-dependent manner. This indicates that the reduction in dietary energy intake from propionate infusion was greater than the energy supplied from infusates and that excess propionate production in the rumen can decrease energy intake in lactating dairy cows consuming highly fermentable diets. However, quadratic effects of propionate infusion were not significant for DMI and ME intake, providing no evidence for a threshold response to infused propionate in feeding behavior and energy intake. As the proportion of propionate in infused VFA increased, meal size tended to decrease and intermeal interval tended to increase in Experiment 1, and meal size decreased and meal frequency tended to decrease in Experiment 2. These observations suggest that propionate plays an important role in feed intake regulation by affecting both satiety and hunger.

ACKNOWLEDGMENTS

We thank R. E. Kreft and D. G. Main for technical assistance.

LITERATURE CITED

1. Allen, M. S. (2000) Effects of diet on short-term regulation of feed intake by lactating dairy cattle. J. Dairy Sci. 83: 1598–1624.

2. Choi, B. R. & Allen, M. S. (1999) Intake regulation by volatile fatty acids and physical fill. S. Afr. J. Anim. Sci. 29: 40–41(abs.).

3. Anil, M. H. & Forbes, J. M. (1980) Feeding in sheep during intraportal

infusions of short-chain fatty acids and the effect of liver denervation. J. Physiol. 298: 407–414.

4. Elliot, J. M., Symonds, H. W. & Pike, B. (1985) Effect on feed intake of infusing sodium propionate or sodium acetate into a mesenteric vein of cattle. J. Dairy Sci. 68: 1165–1170.

5. Farningham, D.A.H. & Whyte, C. C. (1993) The role of propionate and acetate in the control of food intake in sheep. Br. J. Nutr. 70: 37–46.

6. Hurtaud, C., Rulquin, H. & Verite, R. (1993) Effect of infused volatile fatty acids and caseinate on milk composition and coagulation in dairy cows. J. Dairy Sci. 76: 3011–3020.

7. Mbanya, J. N., Anil, M. H. & Forbes, J. M. (1993) The voluntary intake of hay and silage by lactating cows in response to ruminal infusion of acetate or propionate, or both, with or without distension of the rumen by a balloon. Br. J. Nutr. 69: 713–720.

8. Sheperd, A. C. & Combs, D. K. (1998) Long-term effects of acetate and propionate on voluntary feed intake by midlactation cows. J. Dairy Sci. 81: 2240–2250.

9. Wu, Z., Sleiman, F. T., Theurer, C. B., Santos, F., Simas, J. M., Francolin, M. & Huber, J. T. (1994) Effect of isocaloric infusion of glucose in the rumen or propionate in the duodenum. J. Dairy Sci. 77: 1556–1562.

10. Anil, M. H., Mbanya, J. N., Symonds, H. W. & Forbes, J. M. (1993) Responses in the voluntary intake of hay or silage by lactating cows to intraruminal infusions of sodium acetate or sodium propionate, the tonicity of rumen fluid or rumen distension. Br. J. Nutr. 69: 699–712.

11. Deetz, L. E. & Wangsness, P. J. (1981) Influence of intrajugular administration of insulin, glucagon and propionate on voluntary feed intake of sheep. J. Anim. Sci. 53: 427–433.

12. DeJong, A., Steffens, A. B. & De Ruiter, L. (1981) Effects of portal volatile fatty acid infusions on meal patterns and blood composition in goats. Physiol. Behav. 27: 683–689.

13. Quigley, J. D. & Heitmann, R. N. (1991) Effects of propionate infusion and dietary energy on dry matter intake in sheep. J. Anim. Sci. 69: 1178–1187.

14. Allen, M. S. (1997) Relationship between fermentation acid production in the rumen and the requirement for physical effective fiber. J. Dairy Sci. 80: 1447–1462.

15. Dado, R. G. & Allen, M. S. (1993) Continuous computer acquisition of feed and water intake, chewing, reticular motility, and ruminal pH of cattle. J. Dairy Sci. 76: 1589–1600.

16. National Research Council (1989) Nutrient Requirements of Dairy Cattle, 6th rev. ed. National Academy of Sciences, Washington, DC.

17. Van Soest, P. J., Robertson, J. B. & Lewis, B. A. (1991) Methods for dietary fiber, neutral detergent fiber and nonstarch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74: 3583–3597.

18. Hach, C. C., Bowden, B. K., Lopelove, A. B. & Brayton, S. V. (1987) More powerful peroxide Kjeldahl digestion method. J. Assoc. Off. Anal. Chem. 70: 783–787.

19. Karkalas, J. (1985) An improved enzymatic method for the determination of native and modified starch. J. Sci. Food Agric. 36: 1019–1027.

20. American Society of Agricultural Engineers (1968) Method of determining and expressing fineness of feed material by sieving. In: ASAE Standards, S319. ASAE, St. Josph, MI.

21. Grovum, W. L. (1995) Mechanisms explaining the effects of short chain fatty acids on feed intake in ruminants—osmotic pressure, insulin and glucagon. In: Ruminant Physiology: Digestion, Metabolism, Growth and Reproduction (Englehardt, W. v., Leonhard-Marek, S., Breves, G. & Geisecke, D., eds.), pp. 173–197. Ferdinand Enke Verlag, Stuttgart, Germany.

22. Carter, R. R. & Grovum, W. L. (1990) Factors affecting the voluntary intake of food by sheep. 5. The inhibitory effect of hypertonicity in the rumen. Br. J. Nutr. 64: 285–299.

23. DeJong, A. (1982) Patterns of plasma concentrations of insulin and glucagon after intravascular and intraruminal administration of volatile fatty acids in the goat. J. Endocrinol. 92: 357–370.

24. Gonda, H. L., Lindberg, J. E. & Holtenius, K. (1997) Plasma levels of energy metabolites and pancreatic hormones in relation to the level of intake and intraruminal infusions of volatile fatty acids in fed wether sheep. Comp. Biochem. Physiol. 116A: 65–73.

25. Deetz, L. E., Wangsness, P. J., Kavanaugh, J. F. & Griel, L. C., Jr. (1980) Effect of intraportal and continuous intrajugular administration of insulin of feeding in sheep. J. Nutr. 110: 1983–1991.

26. Foster, L. A., Ames, N. K. & Emery, R. S. (1991) Food intake and serum insulin response to intraventricular infusions of insulin and IGF-I. Physiol. Behav. 50: 745–749.

27. McCarthy Jr., R. D., Klusmeyer, T. H., Vicini, J. L., Clark, J. H. & Nelson, D. R. (1989) Effects of source of protein and carbohydrate on ruminal fermentation and passage of nutrients to the small intestine of lactating cows. J. Dairy Sci. 72: 2002–2016.

28. Overton, T. R., Cameron, M. R., Elliot, J. P., Clark, J. H. & Nelson, D. R. (1995) Ruminal fermentation and passage of nutrients to the duodenum of lactating cows fed mixtures of corn and barley. J. Dairy Sci. 78: 1981–1998.

29. SAS Institute Inc. (1990) Segmented model. In: SAS/STAT User's Guide, Version 6, p. 1162. SAS Institute, Cary, NC.

30. Leuvenink, H.G.D., Bleumer, E.J.B., Bongers, L.J.G.M., VanBruchem, J. & VanDerHeide, D. (1997) Effect of short-term propionate infusion on feed intake and blood parameters in sheep. Am. J. Physiol. 272: E997–E1001.

31. Murphy, M. R., Davis, C. L. & McCoy, G. C. (1983) Factors affecting water consumption by Holstein cows in early lactation. J. Dairy Sci. 66: 35–38.