The effect of starch-, fiber-, or sugar-based supplements on nitrogen utilization in grazing dairy cows

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ABSTRACT

Nitrogen utilization in grazing cows is often low due to high concentrations of rapidly soluble and degradable protein in the pasture-based diet. Broadly, opportunities to improve N utilization lie in either reducing the amount of N consumed by the animal, or incorporating more N into milk protein. The goal of this study was to compare the relative importance of dietary N intake and productive N output for improving N utilization in grazing cows fed either starch-, fiber-, or sugar-based supplements. Also, the Cornell Net Carbohydrate and Protein System (CNCPS; Cornell University, Ithaca, NY) was evaluated as a tool to assess cow performance and improve N utilization in pasture-based systems. Eighty-five cows were randomly assigned to 1 of 5 treatments at parturition (17 cows per treatment). Treatments consisted of a pasture-only control and pasture with a starch- (St and StN), fiber- (FbN), or a sugar-based supplement. The StN and FbN treatments contained additional dietary N. Diets were formulated using the CNCPS to supply similar levels of dietary metabolizable energy, but differing levels of dietary N and metabolizable protein. Nitrogen utilization ranged from 22 to 26% across the 5 groups. Cows fed the St diet had the lowest levels of milk urea N, blood urea N, and urinary N excretion and had the highest productive N output (149 g/d). Cows fed the FbN treatment had similar productive N output (137 g/d) and consumed approximately 100 g/d more dietary N than the St treatment, resulting in greater urinary N excretion. Although milk protein yield was moderately greater in the St treatment, quantitatively the difference in N intake (100 g/d) had the greatest effect on N utilization and suggests that controlling dietary N intake should be the first priority when attempting to improve N utilization in grazing cows. No effect was observed of supplementing pasture-fed cattle with sugar on production or N utilization under the conditions of this experiment.

Predictions of metabolizable energy and protein availability for milk yield from the CNCPS were similar to actual milk yield for all treatments. Model-predicted N utilization and excretion reflected the trends observed in the measured data and suggests that the CNCPS can be a useful tool for formulating and evaluating diets to improve N utilization in pasture-based systems.

Key words: modeling, pasture, nitrogen-use efficiency, sustainability

INTRODUCTION

Globally, increased pressure exists to reduce the environmental impact of dairying. Nitrogen management is of particular concern in pasture-based systems due to its impact on water quality in aquifers, rivers, and lakes (Ledgard et al., 1999). Improving N utilization in pastoral systems presents a unique set of challenges, given the requirement for high levels of pasture consumption for low-cost production, the large demands of N by temperate grasses, and the resultant soluble and rapidly degradable nature of pasture protein (Kolver, 2003).

Nitrogen-use efficiency can be broadly defined as the proportion of productive N output from the cow (N accretion, milk N, or N retained by the conceptus), relative to total N consumed (Calsamiglia et al., 2010). An increase in milk protein yield would increase productive N output and improve N utilization for a given N intake. Any N not secreted in milk, or accreted into tissue, will be lost in either feces or urine (Lapierre and Lobley, 2001). Fecal N is relatively fixed (Marini and Van Amburgh, 2003); the greatest opportunity to improve N utilization, therefore, is in either reducing urinary N output, or increasing productive N output (Broderick, 2003; Marini and Van Amburgh, 2003). In pasture-based systems, previous efforts to improve N capture have focused on improving energy supply to the rumen, with the objective of incorporating more ammonia into microbial protein and, thereby, increasing the AA flow to the small intestine (Kolver et al., 1998a; Miller et al., 2001; Sairanen et al., 2005; Moorby et al., 2006). Although important, this assumes that the milk

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yield of the cow is limited by AA supply, which when corrected, should increase milk protein synthesis and secretion. Milk protein synthesis appears more closely linked to milk yield, physiological state, and overall nutritional status than simple substrate availability (Hanigan et al., 2001; Cant et al., 2003). Therefore, in a situation where MP is adequate, increasing AA supply will increase hepatic removal and shift the majority of the associated N back to the urea pool with no real benefit to overall N utilization (Lapierre et al., 2005).

Changing the ratio of starch or sugar to NDF has previously been reported to alter ruminal VFA profiles (Bauman et al., 1971; Beckman and Weiss, 2005), with subsequent effects on milk composition (Broderick, 2003; Beckman and Weiss, 2005). Changing dietary MP and N intake simultaneously tests the effect of substrate supply, dietary N dilution, and their comparative importance in improving N utilization for a given N intake compared with increasing productive N output. The Cornell Net Carbohydrate and Protein System (CNCPS; Cornell University, Ithaca, NY) was previously used to evaluate grazing cows by Kolver et al. (1998b), who reported that the model could realistically predict ME and MP supply and subsequent milk production. Recent changes have been made to improve CNCPS predictions, including a recharacterization of various pool constituents, degradation rates, passage rate assignments (Van Amburgh et al., 2007, 2010), and predictions of N excretion (Higgs et al., 2012). The aim of the current study was to investigate the opportunity to improve productive N output in grazing cows using starch-, fiber-, and sugar-based supplements formulated to supply balanced ME, with differing MP and N intakes. A second goal was to simulate the experimental conditions in the CNCPS and assess its usefulness as a tool to model N utilization under grazing conditions.

MATERIALS AND METHODS

Experimental work was conducted at the DairyNZ Lye Farm (Hamilton, New Zealand; 37°47'S 175°19'E) during July and August 2010. Prior approval for animal use was attained from the Ruakura Animal Ethics Committee (Hamilton, New Zealand).

Experimental Design and Treatments

Eighty-five dairy cows (53 Friesian and 32 Friesian × Jersey; 69 multiparous and 16 primiparous) due to calve over a 21-d period were randomly assigned to 1 of 5 treatments at parturition ($n = 17$); treatments were balanced for milk production (mean of the first 100 DIM from the previous lactation for multiparous cows; 17.7 ± 0.7 kg of milk/cow per day; mean $\pm$ SD),

precalfing BW (549 ± 29 kg), BCS (4.5 ± 0.3 ; 10-point scale; Roche et al., 2004), and age (4.5 ± 0.2 yr).

Dietary treatments consisted of a pasture-only control (CON) and pasture with starch (St and StN), fiber (FbN), or sugar (Sg)-based supplements. The StN and FbN treatments were formulated to supply equal dietary N and MP, whereas the St and Sg treatments had no additional N. A small amount of soybean meal was added to the StN treatment to make it equivalent to the FbN treatment on a true-protein (TP) basis. Corn grain was used as the starch source, wheat middlings as the fiber source, and molasses as the sugar source. Supplements were formulated using the CNCPS v6.1 (Tylutki et al., 2008; Van Amburgh et al., 2010) and fed to support a target of 30 kg of milk. Chemical composition and DMI for each treatment are presented in Table 1. All supplements were offered in pellet form except the Sg treatment, which was in liquid form and fed at a lower rate to prevent adverse health effects. The assumptions used when formulating the supplements were that cows in early lactation (~40 DIM), of similar BW, offered between 30 and 40 kg of ryegrass-based pasture DM/d would consume approximately 15 kg of DM/d (Dalley et al., 1999) and would substitute approximately 0.5 kg of pasture DM/kg of concentrate DM fed (Bargo et al., 2003). Supplements were introduced gradually over a 3-d period and offered in 2 equal portions at a.m. and p.m. milkings. The Sg treatment was provided orally in a diluted bolus (3:1 molasses:water) after each milking. All cows calved in an 18-d period and started their allocated treatment immediately after parturition. The experiment concluded on the same day for all cows meaning the experimental period ranged from 6.5 to 9 wk, depending on the calving date.

Grazing Management

Cows rotationally grazed 37 ha permanently subdivided into 1-ha paddocks (defined grazing area) as one group ($n = 85$). Each paddock was further subdivided using a temporary electric fence to establish grazing conditions that encouraged pasture to be harvested to a postgrazing residual mass of 1,500 to 1600 kg of DM/ha. This has been reported to balance the dual goals of achieving high DMI while maximizing pasture production and quality for future grazing events (Hoogendoorn et al., 1992; Lee et al., 2008). Cows in early lactation have increasing DMI; therefore, pasture allowance was continually reassessed to maintain the target residual pasture mass. Pasture allowance (mean $\pm$ SD) was 29 ± 5 kg of DM/cow per day for the last 3 wk of the study. Pre- and postgrazing compressed sward heights for the same period were 22.9 ± 2.3 and 10.6 ± 1.2 cm, respectively, and pre- and postgrazing

Table 1. Feed intake and chemical composition of experimental diets

Item ¹	Diet ²				
	CON	St	StN	FbN	Sg
Intake (kg/d)					
DMI	11.7	13.8	13.9	15.1	12.6
Ingredient (% of DM)					
Pasture	100.0	72.2	78.4	68.7	86.7
Corn meal	0.0	25.0	16.7	0.0	0.0
Wheat middlings	0.0	2.8	2.2	29.8	0.0
Soybean meal, 48%	0.0	0.0	1.8	0.0	0.0
Fat	0.0	0.0	0.0	1.1	0.0
Urea	0.0	0.0	0.9	0.4	0.0
Molasses	0.0	0.0	0.0	0.0	13.3
Chemical composition (% of DM)					
CP	28.1	23.0	27.3	25.0	25.0
SP (% of CP)	46.0	41.4	42.9	45.7	53.0
ADICP (% of CP)	7.9	6.5	6.8	6.6	7.2
NDICP (% of CP)	33.3	26.9	28.1	26.8	29.3
NDF	39.5	32.6	34.0	38.5	34.3
ADF	22.0	17.5	18.5	19.1	19.1
Lignin (% of NDF)	5.7	8.1	7.2	6.9	13.8
EE	4.9	4.6	4.6	5.7	4.6
Starch	0.3	16.5	11.3	7.1	0.3
Sugar	12.8	10.2	10.9	10.4	21.8
Ash	8.0	6.4	6.8	6.9	8.6

¹SP = soluble protein; ADICP = acid detergent-insoluble CP; NDICP = neutral detergent-insoluble CP; EE = ether extract.

²CON = control (pasture only); St = pasture with a starch-based supplement; StN = pasture with a starch-based supplement and additional N; FbN = pasture with a fiber-based supplement and additional N; Sg = pasture with a sugar-based supplement.

pasture yield was $3,243 \pm 261$ and $1,681 \pm 233$ kg of DM/ha, respectively (mean $\pm$ SD). Measurements were made using a rising plate meter installed with an electronic counter (Farmworks Ltd., Palmerston North, New Zealand). Cows had access to a fresh allocation of pasture twice daily and only returned to the same area when a minimum of 2 leaves had appeared on the majority (>66%) of perennial ryegrass (*Lolium perenne* L.) tillers. The pasture offered consisted of (mean $\pm$ SD) 90.2 (± 2.8)% perennial ryegrass leaf, 2.5 (± 1.4)% perennial ryegrass stem, 1.5 (± 2.2)% white clover (*Trifolium repens*), 0.6 (± 0.7)% weeds (*Sisymbrium officinale*, *Achillea millefolium*, *Taraxacum officinale*, and *Ranunculus sardous*), and 5.2 (± 1.8)% dead material, on a DM basis.

Pasture Measurements

Representative samples of pasture were collected daily by clipping pasture to grazing height from paddocks due to be grazed. Samples were bulked on a weekly basis for the duration of the experiment, and duplicate samples were dried for 48 h at either 100°C for DM analysis or 60°C for analysis of nutrient composition. Samples dried at 60°C were subsequently ground to pass through a 1.0-mm sieve (Christy laboratory mill; Christy Turner Ltd., Suffolk, UK) and analyzed by wet

chemistry for the nutrients required to evaluate the diets in the CNCPS (Tylutki et al., 2008; DairyOne, Ithaca, NY).

Animal Measurements

DMI. Mean group pasture DMI was calculated as the product of the difference between the pre- and postgrazing pasture mass and area grazed daily (Roche et al., 1996). Supplement offered and refused was measured at each milking. Estimations of individual cow pasture DMI were obtained using the *n*-alkane technique outlined by Kennedy et al. (2003). Briefly, each cow was dosed twice daily (at milking) with a capsule containing 356 mg of *n*-dotriacontane (C32; i.e., 712 mg of C32/cow per day) for a 10-d period in wk 6 and 7 of the experiment. Fecal grab samples were collected twice daily from each cow (after milking) during the last 5 d of the 10-d period. Fecal samples from each cow for the 5-d period were bulked and stored at -17°C awaiting alkane analysis. During the same 5-d period, pasture samples were plucked to grazing height, following close observation of the grazing animal, to represent pasture grazed. The *n*-alkane concentration (C25–C36) in pasture, supplement, and feces were determined using gas chromatography (Mayes et al., 1986). The ratio of pasture C33 (tritriacontane) to dosed C32 (*n*-dotria-

contane) was used to estimate pasture DMI. Estimates of daily pasture DMI were calculated as follows:

$$\text{Daily pasture intake (kg of DM/cow)} = \frac{F_i/F_j \times (D_j + I_s \times S_j) - I_s \times S_i}{P_i - (P_j \times F_i/F_j)},$$

where F_i , S_i , and P_i are the concentrations (mg/kg of DM) of the natural odd-chain n -alkane (C33) in feces, supplement, and pasture, respectively; F_j , S_j , and P_j are the concentrations (mg/kg of DM) of the dosed even-chain n -alkane (C32) in feces, supplements, and pasture, respectively; and D_j and I_s are the dose rate (mg/d) of the even-chain n -alkane (C32) and supplement intake, respectively.

Milk and BW. Individual milk yields were recorded daily (GEA Westfalia Separator GmbH, Oelde, Germany). Fat, TP, and lactose concentrations in milk were determined by a MilkoScan FT120 (Foss Electric A/S, Hillerød, Denmark) on a composite from a.m. and p.m. samples collected once (2 consecutive days) each week for the duration of the experiment. Milk composition data were verified by reference techniques for a subset of milk samples (milk fat: Röese-Gottlieb method; IDF, 1987; CP: Kjeldahl techniques; Barbano et al., 1991). Body weight and BCS were measured weekly following the a.m. milking; BCS was assessed on a 10-point scale, where 1 is emaciated and 10 is obese (Roche et al., 2004). These scores can be converted to the 5-point scale of Wildman et al. (1982) using the regression equation generated by Roche et al. (2004; 5-point BCS = $1.5 + 0.32$ 10-point BCS).

Blood. Two 10-mL evacuated blood tubes containing either a sodium heparin pellet (158 IU of sodium heparin) or EDTA (0.117 mL of 15% K_3 EDTA) to prevent coagulation were collected from each cow by coccygeal venipuncture before treatment allocation and weekly thereafter. Plasma was separated ($1,120 \times g$ for 10 min at 4°C) and frozen at -20°C before analysis. Plasma from the EDTA tubes was analyzed for NH_3 concentration (mmol/L), based on the enzymatic kinetic assay described by da Fonseca-Wollheim (1973). Plasma from the sodium heparin tubes were analyzed for NEFA, BHBA, glucose, and urea. Determination of NEFA (mmol/L; colorimetric method using a commercial kit: Wako Pure Chemical Industries Ltd., Osaka, Japan), BHBA (mmol/L; BHBA dehydrogenase assay based on formation of acetoacetate and NADH after addition of NAD), glucose (mmol/L; hexokinase method based on formation of NADPH), and urea (mmol/L; urease hydrolysis method) concentrations were performed on

a Hitachi Modular P800 analyzer (Roche, Basel, Switzerland) at 30°C by Gribbles Veterinary Pathology Ltd. (Hamilton, New Zealand). The inter- and intraassay coefficients of variation were $<2\%$ for all assays.

Urine. Mid-stream urine samples were collected once each week during voluntary urination of cows immediately before the morning milking. After collection, samples were divided into 50-mL aliquots for the analysis of creatinine, urea, uric acid, allantoin, urea, and total N. The aliquots used for the analysis of urea and total N were reduced to $\text{pH} \leq 2$ using approximately 3 mL of 6 mol of hydrochloric acid/L and frozen at -20°C before analysis. Creatinine, uric acid (mmol/L; enzymatic colorimetric assay), and urea (mmol/L; kinetic UV assay) were analyzed using commercial kits (Roche Diagnostic NZ Ltd., Auckland, New Zealand) by Gribbles Veterinary Pathology Ltd. Allantoin was analyzed on a spectrophotometer using a colorimetric assay (Young and Conway, 1942) and total N was analyzed using the Leco total combustion method (Institute of Food, Nutrition and Human Health, Massey University, New Zealand).

CNCPS Inputs

Data used in the CNCPS represented the mean of a 5-d period in wk 7 of the study, coinciding with the n -alkane DMI estimation. Dietary inputs, including DMI, feed ingredients, and the chemical composition of ration are in Table 1. Animal inputs, including milk production, initial BCS, and BW change are presented in Table 2. Other inputs, including stage of lactation, breed, and parity are consistent with the previous description in this section. The contribution of tissue mobilization to predictions of ME and MP milk (Table 2) were estimated from BW change. The chemical composition (fat:protein) of mobilized body reserves changes depending on the BCS of the animal (Fox et al., 1999). To account for this, the composition of reserves mobilized was calculated using the BW change and initial BCS from Table 2 and equations in Fox et al. (2004). Briefly, initial BW and BCS were used to calculate a reference BW at BCS 3 (1–5 scale). Mobilized fat and protein were then estimated using the reference BW and the change from initial to final BW (Fox et al., 2004). Change in BW was preferred to BCS as an estimate of tissue mobilization due to the difficulty in ascertaining small changes in BCS over 1 time period (Ferguson et al., 1994). It was assumed that when MP supply was in excess to requirements, protein mobilized from tissue was used as an energy source and contributed to ME supply.

Table 2. Cornell Net Carbohydrate and Protein System (CNCPS; Cornell University, Ithaca, NY) inputs and predictions for the effect of supplementing different carbohydrate types on N use parameters

Item	Diet ¹				
	CON	St	StN	FbN	Sg
Milk, actual (kg/d)	23	28	26	26	24
ME milk, predicted ² (kg/d)	22	27	27	28	26
MP milk, predicted ² (kg/d)	23	25	26	27	24
Diet-allowable ME milk ³ (kg/d)	7	15	14	15	10
Diet-allowable MP milk ³ (kg/d)	23	24	26	27	24
Initial BW (kg)	441	467	442	448	477
Final BW (kg)	417	449	422	427	453
Initial BCS ⁴	3.8	4.1	3.8	4.0	4.1
Final BCS	3.7	4.0	3.7	4.0	4.0
N intake (g/d)	527	507	607	604	504
Productive N (g/d)	116	149	134	137	115
Fecal N (g/d)	155	163	182	187	155
Urine N (g/d)	303	230	331	320	279
N balance (g/d)	-48	-34	-39	-40	-45
MP intake (g/d)	1,548	1,697	1,776	1,889	1,547
MP from bacteria (% of MP intake)	34	42	38	38	40
ME intake (MJ/d)	123	154	151	159	136
Productive N:urine N	0.38	0.65	0.40	0.43	0.41
Productive N:intake N	0.22	0.29	0.22	0.23	0.23
Milk TP:MP supply ⁵	0.48	0.56	0.48	0.46	0.47

¹CON = control (pasture only); St = pasture with a starch-based supplement; StN = pasture with a starch-based supplement and additional N; FbN = pasture with a fiber-based supplement and additional N; Sg = pasture with a sugar-based supplement.

²Includes contributions from body reserves.

³ME and MP milk refer to the potential milk that could be produced at the predicted supply of metabolizable energy and metabolizable protein (ME and MP) by the CNCPS.

⁴Measured on a 1 to 10 scale (Roche et al., 2004).

⁵TP = true protein.

Statistical Analysis

Data are expressed as means of the last 3 wk of the study and were analyzed using a restricted maximum likelihood model (REML) in GenStat 13.2 (VSN International, 2010). The model included the fixed effects of calving group (3 groups to account for calving date), age (primiparous and multiparous), week of study, treatment, and the interaction of calving group and week. The effects of calving group, parity, and week of study were included to account for nontreatment variation and are not considered important in explaining treatment effects. Cow was included as a random effect. Treatment effects were considered significant at $P < 0.05$. The least significant difference for the error degrees of freedom was approximately $2 \times$ the standard error of the difference.

RESULTS

Animal Observations

The type of supplement fed to cattle on treatment affected milk yield ($P < 0.01$), yield of TP ($P < 0.001$)

and lactose ($P < 0.01$), but not milk fat (Table 3). Cows fed the St diet had the highest milk and TP yields ($P < 0.01$), followed by those fed the FbN and StN diets. Cows fed the Sg diet had similar milk and milk components to cows fed the CON diet. Concentrations of fat, TP, and lactose were affected ($P < 0.01$) by treatment (Table 3). Milk fat concentration was lower in cows fed the St supplement than in cows in the other 4 groups, which did not differ from each other. True protein concentration was greater ($P < 0.01$) in the St treatment compared with the CON and Sg treatments, but similar to the StN and FbN treatments. Milk urea N concentration was lower in cows fed the St diet ($P < 0.001$) compared with all other diets.

Cows fed the St diet had more than 1 mmol/L less urea in blood than the other treatments, which was consistent with the ratios of urinary urea to creatinine and urinary N to creatinine (Table 4). There were no differences in the ratio of purine derivatives (PD) to creatinine. Blood concentrations of BHBA were elevated in CON and Sg cows, but similar among the other treatments. Blood NEFA concentrations were greater ($P < 0.05$) in cows fed the Sg and FbN treatments, but similar among the other treatments.

Table 3. Effects of supplementing different carbohydrate types to grazing dairy cows in early lactation on milk yield and milk composition

Item	Diet ¹					SED ²	P-value ³
	CON	St	StN	FbN	Sg		
Yield (kg/d)							
Milk	23.1	27.7	25.5	26.2	23.6	1.34	0.005
Fat	1.03	1.07	1.11	1.16	1.06	0.061	0.33
TP ⁴	0.74	0.95	0.85	0.87	0.73	0.038	<0.001
Lactose	1.13	1.38	1.25	1.28	1.14	0.063	0.001
Milk composition (%)							
Fat	4.44	3.88	4.39	4.41	4.57	0.209	0.016
TP	3.20	3.43	3.34	3.34	3.11	0.075	0.001
Lactose	4.89	4.99	4.91	4.92	4.85	0.037	0.010
MUN (mmol/L)	7.24	5.10	7.09	6.40	6.60	0.228	<0.001

¹CON = control (pasture only); St = pasture with a starch-based supplement; StN = pasture with a starch-based supplement and additional N; FbN = pasture with a fiber-based supplement and additional N; Sg = pasture with a sugar-based supplement.

²Standard error of the difference.

³Refers to the overall treatment effect. The least significant difference for this study was $2 \times \text{SED}$. Therefore, individual treatment means were considered significantly different when they differed by $>2 \times \text{SED}$.

⁴TP = true protein.

CNCPS Predictions

Predictions from the CNCPS are presented in Table 2. Metabolizable energy and MP intake was similar among the St and FbN treatments and Sg and StN treatments, respectively, but predicted MP allowable milk was considerably higher than ME allowable milk. Cows fed the StN and FbN diets were similar in total N intake (~ 600 g of N/d), which was approximately 100 g greater than for those fed the St and Sg diets and 70 g greater than the CON. When tissue mobilization was included in the model, predicted ME and MP allowable milk were balanced and similar to actual milk production.

Predicted urinary N excretion followed the trends evident in ratios of urinary N and urinary urea to creatinine (Table 4). The ratios of productive N:urinary N, productive N:intake N, and milk TP:MP supply were all greater in cows fed the St diet, but similar among those fed the other diets.

DISCUSSION

Efficiency of N utilization in dairy cows is typically low, averaging around 25%, but can range from 15 to 40% (Calsamiglia et al., 2010). Trends evaluated over a wide range of dietary and management conditions

Table 4. Effects of supplementing different carbohydrate types to grazing dairy cows in early lactation on parameters of N and energy metabolism

Item	Diet ¹					SED ²	P-value ³
	CON	St	StN	FbN	Sg		
Blood parameter (mmol/L)							
BUN	7.05	5.10	6.72	6.07	6.60	0.227	<0.001
Glucose	4.05	4.20	4.12	4.34	3.97	0.093	0.002
NEFA	0.54	0.54	0.52	0.63	0.65	0.052	0.036
BHBA ⁴	0.62 (−0.21)	0.38 (−0.42)	0.48 (−0.32)	0.42 (−0.37)	0.65 (−0.19)	−0.041	<0.001
Urine parameter ⁴							
PD:creatinine ⁵	2.97 (0.47)	2.86 (0.46)	2.49 (0.40)	3.09 (0.49)	2.49 (0.40)	−0.045	0.14
N:creatinine	0.23 (−0.65)	0.16 (−0.79)	0.19 (−0.71)	0.21 (−0.68)	0.20 (−0.70)	−0.039	0.004
Urea:creatinine	78.44 (1.89)	54.63 (1.74)	75.40 (1.88)	73.16 (1.86)	69.27 (1.84)	−0.025	<0.001

¹CON = control (pasture only); St = pasture with a starch-based supplement; StN = pasture with a starch-based supplement and additional N; FbN = pasture with a fiber-based supplement and additional N; Sg = pasture with a sugar-based supplement.

²Standard error of the difference.

³Refers to the overall treatment effect. The least significant difference for this study was $2 \times \text{SED}$. Therefore, individual treatment means were considered significantly different when they differed by $>2 \times \text{SED}$.

⁴Data were logarithm transformed for statistical analysis. Numbers outside the parentheses are back-transformed values and numbers inside the parentheses are logarithm transformed. The SED corresponds to the transformed values.

⁵PD = purine derivatives (allantoin + uric acid).

indicate that dietary CP concentration is the most important factor influencing the efficiency of N use (Huhtanen and Hristov, 2009). Efficiencies as high as 43% (Frank and Swensson, 2002) and 37% (Noftsker and St-Pierre, 2003) have been reported in the literature in TMR-fed cows and as high as 38% in cows fed ryegrass-based diets (Moorby et al., 2006). The observed N efficiencies for cows in the current study ranged from 22 to 29% (Table 2), which, according to Calsamiglia et al. (2010), would be classified as ranging from low to moderately high, respectively. Cows with the highest N-use efficiency in the current study (St) still wasted 10% more N than cows in the study of Moorby et al. (2006). The major difference between the 2 studies was the CP content of the pasture, which was 28.1% in the current study (Table 1) compared with approximately 10% in the study of Moorby et al. (2006). Attempts have been made to improve the retention of dietary N by synchronizing the supply of energy and protein in the rumen through supplementation (Kolver et al., 1998a), and also through feeding pasture cultivars bred to have higher sugar content (Edwards et al., 2007). Effects have generally been transient, with no real improvement in N utilization, suggesting that N intake is a more important factor in improving N utilization (Henning et al., 1993; Kolver et al., 1998a; Kim et al., 1999; Edwards et al., 2007).

Pasture CP in the current study was higher than anticipated (Table 1); this resulted in predicted MP-allowable milk being approximately 10 kg higher than ME-allowable milk (ME- and MP-allowable milk refer to the potential milk that could be produced at the predicted supply of ME and MP by the CNCPS; Table 2). Unfortunately, the StN treatment was less palatable than the other treatments and resulted in lower-than-formulated ME intake. Despite this, N intake and predicted MP-allowable milk were similar among cows fed the St and Sg treatments and StN and FbN treatments (Table 2) allowing the comparison of both dietary N dilutions and the effect of carbohydrate type. Compared with the CON, cows fed the Sg diet had a lower ratio of urinary urea:creatinine and MUN; however, urinary N:creatinine and BUN were similar. This suggests a small N dilution effect consistent with the reduction in dietary N intake (Table 2), but no improvements in the overall efficiency of N use. In contrast, cows fed the St diet consumed the same amount of dietary N as those fed the Sg diet, but had a 0.06-U greater N-use efficiency (Table 2). Cows fed the St diet consumed more ME than those fed the Sg diet. However, cows fed the Sg diet also consumed more ME than the CON and no difference in production existed between these 2 treatments. Supplementing with sucrose has previously been reported to decrease rumen pH and decrease the rate of

NDF digestion (Huhtanen and Khalili, 1991; Chamberlain et al., 1993). In the experiment of Huhtanen and Khalili (1991), cows were fed 1 kg of DM/d of sucrose, which reduced the rate of NDF digestion by 1.5%/h. Pasture NDF in the current study was calculated to digest at a rate of 7%/h (Van Amburgh et al., 2003). Reducing this digestion rate from 7 to 5.5%/h in the CNCPS reduced the ME-allowable milk from 26 to 24 kg and probably explains the lack of response in the Sg treatment. Pasture in the current study was 12.8% sugar and the administration of molasses increased the sugar content in the diet of the Sg cows to 21.8% (Table 1). No difference was observed in the ratio of PD:creatinine, suggesting that total microbial growth was not changed by treatment (Valadares et al., 1999), although a tendency was observed for PD:creatinine to be lower in Sg and StN cows. Therefore, supplementing molasses to cows consuming high-quality spring pasture using the method used in the current experiment (1.5 kg/d split in 2 feeds at milking) has no production benefit.

The effect of supplementing a fermentable fiber source was also investigated (FbN) and compared with supplementing starch (StN and St). Cows fed the FbN diet had lower BUN and MUN concentrations than cows fed the StN diet; however, no differences existed in N excretion (Table 4) or predicted N utilization (Table 2). Numerically, cows fed the St diet produced more milk protein than cows fed the FbN diet. Rius et al. (2010) reported increased milk protein synthesis with the addition of postruminal starch, a result, most likely, of increased concentrations of insulin and IGF-1 (Griinari et al., 1997; Mackle et al., 1999; Rius et al., 2010). In the current study, differences in N excretion among treatments could be largely explained by differences in N intake (Table 2), with only subtle differences in productive N when ME intake was similar. When comparing the St and FbN treatments, approximately 90% of the difference in N excretion could be attributed to a reduction in N intake, whereas 10% could be attributed to higher productive N output. These findings are in agreement with the conclusion of Huhtanen and Hristov (2009) that reducing N intake is the most important factor in reducing N losses from dairy operations.

Predictions from the CNCPS suggest that cows were consuming adequate dietary MP at the given level of milk, but dietary ME was limited; this is similar to the results of Kolver et al. (1998b). A major difficulty when conducting grazing studies is accurately estimating DMI (Bargo et al., 2003). Intakes reported in this study (Table 2) are based on the *n*-alkane technique described by Kennedy et al. (2003). The dosing and sampling period for this assay coincided with a period

of prolonged wet weather, which reduced pasture utilization and probably explains the low pasture DMI (Holmes et al., 2002). However, pasture intake could also have been underestimated if the recovery of dosed C32 *n*-alkane was lower than expected (Kennedy et al. 2003). Blood NEFA increased sharply and BW decreased sharply over this period (data not presented), which is consistent with the ME deficit predicted by the CNCPS. The model predicted cows fed the Sg and the CON diets to be most limited in dietary ME intake compared with milk produced, which is consistent with higher concentrations of BHBA (Table 4). Mobilization of tissue accounted for 36% of the ME requirement for cows fed the CON diet, which is comparable to levels reported from cows in the first 3 wk of lactation, but are high for cows in the fifth week of lactation (Komaragiri and Erdman, 1997; Pedernera et al., 2008). Including the recorded change in BW into the CNCPS aligned predicted ME and MP milk closely with actual milk for all treatments (Table 2) and is consistent with previous evaluations of the CNCPS under grazing conditions (Kolwer et al., 1998b). Predictions of N excretion reflect measured N excretion (Table 4). Cows fed the St diet were predicted to excrete 70 g/d less urinary N than the CON and approximately 100 g/d less than cows fed the StN and FbN diets, respectively. Although 70 g/d may seem inconsequential, in a herd of 1,000 cows, this reduction is equivalent to 1,000 kg less urea excreted per week and, if sustained, represents a considerable reduction in N loss to the environment. Similar effects could be achieved if CP levels in pasture were reduced (Moorby et al., 2006). Given that only 1 data point was modeled per treatment, care must be taken when interpreting these results. However, model predictions were consistent with the recorded data and suggest that the CNCPS can be successfully used as a tool to formulate diets to improve N utilization in grazing cows.

CONCLUSIONS

Nitrogen utilization can be improved by including high-energy, low-protein supplements into the diets of grazing dairy cows. Reducing dietary N intake appears to be the most important factor in improving N utilization when ME intake is the same. However, subtle improvements in milk protein output can be achieved by feeding starch-, compared with fiber- or sugar-based supplements. Feeding additional sugar to cows fed high-quality spring pasture had no real benefit in the current study. Predictions from the CNCPS were consistent with measured data and predicted ME- and MP-allowable milk were close to measured milk production when estimations of tissue mobilization were included. Predictions of N utilization also reflected the

measured data, indicating that the CNCPS is a useful tool in formulating diets to reduce N losses to the environment.

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