Feeding birdsfoot trefoil (*Lotus corniculatus*) reduces the environmental impacts of dairy farming

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ABSTRACT

An indoor feeding experiment using 16 late lactation Friesian dairy cows measured effects of increased proportions of birdsfoot trefoil (*Lotus corniculatus*; Lotus) in the diet on the partitioning of nitrogen (N) between milk, faeces and urine. Cows were housed in metabolism stalls for 12 days and fed diets containing 0%, 15%, 29% or 45% Lotus (% Lotus dry matter (DM) per DM intake (DMI)) with the remainder perennial ryegrass pasture. Increasing the proportion of Lotus in the diet did not affect DMI but increased milk (P <0.001) and milksolids yields (P <0.01). The percentage of N intake partitioned to milk increased from 16% (0% Lotus) to 21% (45% Lotus) (P <0.01) with an increasing proportion of Lotus in the diet. The percentage of N intake excreted as faecal N also increased from 29% (0% Lotus) to 38% (45% Lotus) (P <0.001) while the percentage excreted as urinary N was reduced from 49% (0% Lotus) to 34% (45% Lotus) (P <0.01). Partitioning of N towards faeces has a favourable environmental impact since faecal N is largely retained in soils whereas urinary N is subject to volatilisation as ammonia, losses as nitrous oxides (a greenhouse gas) and leaching into groundwater.

Keywords: birdsfoot trefoil; dairying; environment; lotus; nitrogen.

INTRODUCTION

The drive for environmental sustainability, whilst increasing dairy production and profitability, poses challenges for farmers. Recent increases in milk production have been associated with increased pasture growth, especially in early spring and autumn, increasing the lactation length (Holmes, 2007). These developments have been partially dependent upon large applications of nitrogenous fertiliser (McGrath *et al*., 1998). The use of nitrogenous fertiliser to promote grass growth has resulted in dietary crude protein (CP; nitrogen (N) x 6.25) concentrations often as high as 30% of dry matter (DM) (Moller, 1997), exceeding requirements of 18 to 20% CP for cows producing 20 to 35 kg milk/d (Kolver, 2000).

Undigested feed N is excreted in the faeces, but most of the N which is absorbed and not incorporated into milk or liveweight (LW) gain, is excreted in the urine. Urinary N is responsible for about 60% of nitrous oxide (N_2O) emissions from agriculture (de Klein & Ledgard, 2001) and makes a substantial contribution to leachate in groundwater, lakes and rivers (Ledgard *et al*., 2000). N excreted in urine also represents a transfer of forage N to a small concentrated patch that promotes grass growth but is avoided by cattle, so reducing feed supply and increasing dead matter accumulation in the patches (Pacheco & Waghorn, 2008).

Options for reducing urinary N excretion include feeding diets with a lower CP concentration, the preferred option, or diverting dietary N away from urine, to faeces. Condensed tannins (CT) present in lotus species are able to divert dietary N from urine to faeces in sheep (Waghorn *et al*., 1994) while reducing methane emissions and increasing milksolids production in cattle (Woodward *et al*., 2004).

Unfortunately, forages containing CT do not have a major role in New Zealand farming because appropriate management practices have not been developed and DM yields from lotus species are lower than from ryegrass-dominant pasture (Waghorn *et al*., 1998). However, requirements to mitigate greenhouse gas (GHG) emissions and improve sustainability of dairy farming suggests CT could have an important role in animal feeding if they could be introduced in a cost effective way. This experiment was undertaken to measure effects of incremental substitution of ryegrass pasture with birdsfoot trefoil (*Lotus corniculatus* (Lotus)) on dry matter intake (DMI), production and the partitioning of dietary N between milk, faeces and urine in lactating cows.

MATERIALS AND METHODS

Experimental design and management

A 12-day experiment was conducted at DairyNZ's Lye Farm, Hamilton, in March 2007 using 16 multiparous Holstein-Friesian dairy cows in late lactation (218 ± 17 days in milk). It was intended that cows would receive 0%, 25%, 50% or 75% Lotus DM in the diet with the remainder perennial ryegrass-based pasture. However, botanical measurements showed the Lotus contained approximately 40% other species so the four treatments were actually 0%, 15%, 29% and 45% Lotus DM. Cows were allocated to the four treatments on the basis of breeding worth and production worth indices, and pre-experiment milk yield and live weight (LW) measurements when all cows grazed perennial ryegrass-based pasture.

For the first three days of the experiment, cows allocated to Lotus treatments were given appropriate amounts of fresh cut Lotus in a barn after morning milking to accustom them to the diets. Once the Lotus was eaten in 2 to 3 hours the cows were released outside to graze ryegrass-based pasture together with the 0% Lotus cows. On the fourth day, all cows were brought into individual metabolism stalls and fitted with faeces and urine collection apparatus. Freshly cut ryegrass pasture and Lotus were mixed in appropriate proportions and the cows were fed diets containing either 0%, 15%, 29% or 45% Lotus DM at 09:00 h and 17:00 h daily to ensure daily refusals were 10 to 15% of feed offered. Measurements presented here were based on a five-day digestion and balance period during the last six days of the trial.

TABLE 1: Chemical and botanical composition of the treatment diets containing increasing amounts of Lotus. Chemical composition values are for bulked samples collected during the five-day measurement period. Botanical composition values are the daily means for each treatment collected during the five-day measurement period

Component	Proportion of Lotus in diet (%)			
	0	15	29	45
Chemical composition				
Dry matter (%)	15.8	16.0	16.1	16.3
Crude protein (g/100g DM)	22.6	22.5	23.3	23.6
Nitrogen (g/100g DM	3.62	3.60	3.73	3.78
Lipid (g/100g DM)	3.4	3.3	3.3	3.3
Acid detergent fibre (g/100g DM)	26.7	26.3	26.0	24.9
Neutral detergent fibre (g/100g DM)	45.9	45.8	43.6	42.1
Soluble sugars and starch (g/100g DM)	10.8	7.4	8.3	4.8
Metabolisable energy (MJ/kg DM)	11.3	11.0	11.0	11.5
Total condensed tannin (g/100g DM)	0.00	0.65	1.26	1.91
Botanical composition				
Ryegrass and other grasses (g/100g DM)	76.3	61.4	47.4	31.9
Lotus (g/100g DM)	0.3	15.3	29.3	45.1
White clover and other legumes (g/100g DM)	14.8	11.8	8.8	5.7
Weeds (g/100g DM)	1.4	5.8	10.1	14.4
Dead (g/100g DM)	7.2	5.7	4.4	2.9

TABLE 2: Intake, milk production and milk composition data from cows fed diets containing increased amounts of Lotus. Values are the means for each treatment during the five-day measurement period. SED = Standard error of difference, N = Nitrogen.

Component	Proportion of Lotus in diet (%)				
	0	15	29	45	SED
Intake (kg DM/cow/d)	15.1	15.2	15.1	14.7	0.3
Milk yield (kg/cow/d)	14.3	15.4	17.0	18.5	1.1
Milk fat (%)	5.29	5.13	4.97	4.90	0.28
Milk protein (%)	3.60	3.74	3.71	3.69	0.16
Milk solids yield (kg/cow/d)	1.27	1.37	1.47	1.59	0.11
Milk N output (g N/cow/d)	80.7	90.6	99.1	106.9	6.8

Intake and feed measurments

Individual DMI was calculated from the fresh weights and DM content (after drying for 48 h at 95^{o}C) of feed offered and refused. The botanical composition of the feed offered and, in particular, the proportion of Lotus in the respective treatment diets were also determined daily. Bulked samples of feed offered during measurement period were oven dried at 65^{o}C and the chemical composition, including total CT concentration, measured using near infrared spectrophotometry (NIRS systems 6500; FeedTECH). The results were used to calculate the N concentration (%) in the diets (crude protein (%) ÷ 6.25) and total N intake (g/d) of each cow.

Milk measurements

Milk yield (kg/cow/d) was measured daily (evening + morning) and samples taken to determine composition of milkfat (%) and milk protein (%) using a Milkoscan 133B analyser (Foss Electric, Denmark). The N concentration (%) of the individual milk samples was then calculated (milk protein (%) ÷ 6.38).

Faeces and urine measurements

Total faeces and urine output (kg/cow/d) were collected daily into separate containers, with the urine containers sealed to minimise N volatilisation losses. Faeces samples were freeze dried and urine samples acidified before measurement of N concentration (%) by total combustion method (AOAC Official Method 968.06, LECO TruSpec CN instrument).

N partitioning calculations

Total N outputs (g/d) were calculated daily for each cow from the sum of N

TABLE 3: Feed nitrogen (N) intake and faecal and urinary N concentrations and output measurements from groups of cows fed increasing amounts of Lotus in their diets. Values are the means for each treatment during the five-day measurement period. Calculations of the partitioning of intake N to milk, faeces and urine as well as that retained are also given as means for each treatment during the five-day measurement period. SED = Standard error of difference, MS = Milk solids.

Component	Proportion of Lotus in diet (%)				
	0	15	29	45	SED
Feed N intake (g N/cow/d)	496	505	511	510	10
Faecal N concentration (%)	2.68	2.85	3.24	3.41	0.07
Faecal N output (g N/cow/d)	144	160	182	194	6
Urine N concentration (%)	0.65	0.62	0.50	0.46	0.04
Urinary N output (g N/cow/d)	244	214	193	174	23
Urinary N output (g N/kg MS)	194	158	133	110	22
Distribution of dietary N intake (%)					
Milk	16.3	18.0	19.4	21.0	1.5
Faeces	29.1	31.7	35.6	37.9	1.5
Urine	49.0	42.6	37.8	34.2	4.1
Retained	5.6	7.7	7.2	6.9	4.4

partitioned to milk, faeces and urine. Retained N was calculated from N intake minus total N output. The distribution of N between milk, faeces and urine was expressed as a percentage of the total N intake.

Statistical analyses

Data from the five measurement days were averaged for individual cows and these means were analysed using analysis of variance (Payne, 2007). Milk yield pre-trial was included as a covariate for the milk production data. Linear and quadratic contrasts of treatment were included in the analyses as fixed effects to test the influence of increasing the level of Lotus in the diet. The quadratic contrasts were not significant for any of the variables.

RESULTS

Feed characteristics

The Lotus forage contained 60% of DM as Lotus with the remainder white clover, weeds and grasses (Table 1). The Lotus forage was good quality and contained 26.8 % CP, 40.5 % neutral detergent fibre (NDF), and 2.51 % CT in the DM, with an estimated metabolisable energy (ME) content of 11.8 MJ/kg DM. The ryegrass pasture had a similar chemical composition and contained 22.6 % CP, 45.9 % NDF and 11.3 MJ ME/kg DM. As a result all diets had a similar chemical composition with dietary CT concentrations ranging from 0 to 1.91% of the DM (Table 1).

DMI and milk production

Increasing the amount of Lotus in the diet did not significantly affect DMI or N intakes (Table 2). Increased Lotus resulted in higher daily milk ($P < 0.001$) and milksolids yields ($P < 0.01$). Cows fed 45% Lotus produced 30% more milk and 25% more milksolids than cows fed 0% Lotus. There were no differences in milkfat and protein concentration (Table 2).

N partitioning

The N intake and total N output of the cows did not change with increasing Lotus in the diet (Table 2). The proportion of N intake partitioned to milk increased (Table 3) from 16% to 21% as Lotus increased from 0% to 45% ($P < 0.01$) due to the increase in milk yield (Table 2). The N concentration in the faeces was higher from cows on the 45% Lotus treatment compared with those on the 0% Lotus treatment ($P < 0.0001$), while the urinary N concentration decreased ($P < 0.001$) as the Lotus content of the diet increased (Table 3). As a result, the proportion of N intake partitioned to faecal N increased from 29% (0% Lotus) to 38% (45% Lotus) ($P < 0.001$) and the proportion excreted as urinary N was reduced from 49% (0% Lotus) to 34% (45% Lotus) ($P < 0.01$) (Table 3).

DISCUSSION

The forages offered in this experiment were high quality, with DM contents averaging 17.3% for the Lotus and 15.8% for ryegrass. Because the CP concentration of both Lotus and ryegrass forages were similar there was little difference in CP concentration between the treatment diets, enabling the effects of substitution with Lotus to be interpreted without confounding effects of changing dietary CP concentration. Experiments with sheep show that CT lowers proteolysis in the rumen (McNabb *et al*., 1996) and the binding between CT and protein at neutral pH in the rumen slows the rate of protein absorption through the small intestine (Wang *et al*., 1996).

The net effect of dietary CT is a reduction in ammonia released from rumen digestion and increased flow of plant protein to the intestine for absorption with an increased concentration of N in faeces and reduced urinary N output (Waghorn, 2008).

Treatment N intakes ranged from 496 to 510 g/cow/d and there was a linear increase in faecal N excretion from 144 to 194 g/cow/d ($R^2 = 0.98$) and a linear decrease in urinary N excretion from 244 to 174 g/cow/d ($R^2 = 0.98$) as the proportion of Lotus in the diet increased (Table 3). Urinary N output expressed in terms of milksolids yield was 43% lower from cows fed 45% Lotus compared with cows fed the 0% Lotus treatment (Table 3). Applied to a typical New Zealand dairy herd of 320 cows, 45% Lotus fed with pasture would reduce urinary N from 78 to 56 kg/d. The difference is equivalent to 50 kg less urea deposited from one day's intake of about 4.8 tonnes of DM for the herd. Effects would be greater in early lactation when intakes are higher, but responses to higher dietary N concentration are not known.

Summation of N in milk, faeces and urine (Table 3) accounted for 92 to 94% of N intake. The unaccounted N was 27 (0% Lotus) to 39 (15% Lotus) g/d and was associated with accretion in body tissue gain, conceptus growth, losses to volatilisation and errors in measurement of feed eaten, urine, faecal and milk N outputs.

Diversion of N from urine to faeces will retain N in soils, because losses from faeces appear to be minor (Ledgard *et al*., 2000), especially with CT in the diet. In contrast, major losses from urinary N include leachate, N_2O and ammonia. Leached nitrate from dairying is predominantly from urine patches (Ledgard *et al*., 2000). N concentration in urine patches is equivalent to 670 to 1,110 kg N/ha (Verloop *et al*., 2006) and nitrate leaching is 30 to 60 kg N/ha/yr as fertiliser N application reached 200 kg N/ha/yr (Ledgard *et al*., 2000).

About 60% of N_2O from pastoral grazing comes from urine patches, and N_2O emissions from agriculture account for about 16% of the New Zealand greenhouse gas inventory (New Zealand Climate Change Office, 2003). Although emissions are affected by soil and climatic conditions, de Klein *et al*. (2008) calculated values from cattle urine to be 1.5% of urinary N input compared with 0.2% of N in dung.

In addition to benefits associated with partitioning excess N away from urine toward faeces, increasing the proportion of Lotus in the diet increased milksolids production (R^2 = 0.99). Research with Lotus fed as a sole diet to lactating dairy cows has demonstrated substantial benefits for milk production. Woodward *et al*. (1999) showed 42% of the gains were attributable to the CT, and the remainder was a 'legume' effect. A 13% reduction in ruminal methane production (g/kg DMI) occurred when cows were fed Lotus compared to ryegrass-dominant pasture with 66% of this reduction due to the effects of CT (Woodward *et al*., 2004). Experiments with sheep and cattle have shown similar reductions in methane emissions attributable to CT, despite several sources and concentrations of CT in the diet (Waghorn *et al*., 2002; C. Grainger, Personal communication).

CONCLUSION

The ability of Lotus to alter the partitioning of N in cattle between faeces and urine has been demonstrated. Previous research with sheep has shown the effects were due to the CT in the Lotus. In this study even small proportions of Lotus in the diet of 15% Lotus with only 0.65% CT in the DM, affected the partitioning of N. Increasing CT up to 1.91% of the DM progressively increased the impact. The poor yield of Lotus compared to ryegrass limits its potential for inclusion in dairy systems. However, a more competitive cultivar (Sivakumaran *et al*., 2006) or one with a higher level of CT or more astringent CT (Waghorn, 2008) may enable smaller quantities to be fed with a similar reduction in urinary N to that reported here. Additional benefits are likely to include a reduction in methanogenesis and increased milksolids yield. If farmers received an incentive to lower methane and N_2O by dietary means, Lotus could form a major component of future diets for dairy cows.

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