The effects of substituting autumn nitrogen fertiliser with gibberellic acid on feed supply and milk production

ME Miller*, GR Edwards, H Hague and RH Bryant

Faculty of Agriculture and Sciences, PO Box 84, Lincoln University, Lincoln 7647, Christchurch, New Zealand

**Corresponding author*. Email: melanie.miller@lincolnuni.ac.nz

Abstract

The aim of this study was to investigate the use of gibberellic acid (GA) to improve feed supply and milk yield in late lactation. A grazing study at Lincoln used 36 lactating, mixed-age, Friesian-Jersey cross dairy cows balanced over three un-replicated treatments of perennial-ryegrass and white-clover pasture treated with nitrogen (N) fertiliser as urea (50 kg N/ha), GA as ProGibb® SG (8 g GA/ha) or untreated (Control). Treatments were applied in late March 2014 and grazed over 12 days in May. There was no effect of treatment on herbage pre-grazing pasture mass (2058±6.9 kg DM/ha, P=0.20), clover (3.6±0.86%, P=0.53), ryegrass (88.0±2.33%, P=0.24) or crude protein (CP) content (17.8±1.60%, P=0.31). All milk yield components: 14.6±0.40 L/day, 1.4±0.09 kg MS/cow/day, 5.4±0.33% fat and 4.4±0.21% protein were similar among treatment groups (P=0.71, 0.14, 0.16 and 0.06, respectively). Although faecal-N content of cows on GA pasture (3.72% N) was higher (P<0.001) than cows on N-fertilised pasture (3.45% N) there was no treatment effect (P>0.05) on urinary-N content (0.4 ± 0.02% N) or N excretion in milk (101±4.4 g N/cow/d). The lack of response to GA or N fertiliser highlights the importance of timing of growth promotants for late lactation.

Keywords: perennial ryegrass; white clover; urea; growth promotants; dairy cow; milk production

Introduction

Late-season pasture growth drives lactation length as farms consider animal condition and pasture covers leading up to winter for their drying-off decisions. New Zealand dairy pastures are typically a combination of perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*) that provide a primary feed source to meet seasonal feed demands. Application of N-based fertiliser is a standard practice used, throughout the season, to maximise pasture production. Over the past two decades, with increases in herd size and stocking rate, the demand for pasture has increased and, with this, the amount of N fertiliser used within New Zealand has more than doubled to a rate of over 400,000 tonnes N/year (New Zealand Fertiliser Association) of which 63% is used in the dairy industry. High rates of fertiliser N and urinary-N are important sources for N loss (Ledgard et al. 1997) and the impact of this on the environment, particularly nitrate leaching, on water quality has been of growing concern (Ledgard et al. 1999, WHO 2011). Urinary-N deposited in autumn presents a high risk of N leaching due to low plant N uptake and high drainage during winter (Selbie et al. 2014). While mitigation strategies continue to be investigated (de Klein et al. 2009; Whitehead et al. 2015; Bryant et al. 2019) a key driver to reduce N surplus is to reduce N inputs. To regulate N fertiliser use on farms, the government has imposed N-caps which will have considerable impacts on the feed supply of N-reliant farms. An alternative option for managing feed supply in the absence of N fertiliser is the use of GA. GA, which is a naturally occurring growth hormone, stimulates cell elongation, breaks dormancy and, when applied exogenously, can stimulate out-of-season dry-matter (DM) production (Matthews et al. 2009). Pasture response to low rates of exogenous GA (8-10 g GA/ha) include increased DM production in late winter, early spring (Bryant et al. 2016; Zaman et al. 2014) and autumn (van Rossum et al. 2013; Zaman et al. 2014) and reduction in herbage protein percent (Bryant et al. 2016). While these responses, which have been observed in small plot studies, offer an opportunity for farmers to reduce N intake and maintain milk yield, there has been no paddock-scale studies to test these hypotheses. This study aimed to determine the impact of using GA in late lactation on pasture production, milk yield and N-use efficiency for milk production.

Materials and method

Site and design

A grazing trial was carried out on the Lincoln University Research Dairy Farm (43°38'S, 172°28'E; 17 m above sea level) between March and May 2014 with approval from the Lincoln University Animal Ethics Committee (#567). The soil type is a Templeton fine sandy loam. The pasture is a tetraploid perennial ryegrass (cv. Bealy, AR37) and white clover (cv. Kopu II) pasture, sown in 2011. The area had been irrigated with effluent during non-grazing periods. The six-hectare experimental area consisted of 4 × 1.5 ha paddocks. The experiment was a completely randomised design consisting of three treatments: no fertiliser or GA (Control), N fertiliser (urea, Ravensdown) and GA (ProGibb® SG, Nufarm 2016).

Treatment application and pasture preparation

Four weeks prior to the experiment (27 March 2014), each paddock was grazed by dairy cows over several days, followed by mowing to a uniform compressed height of approximately 3.5 cm. Within three days of mowing, N fertiliser was applied as granules at a rate of 50 kg N/ha as urea (46% N) while the GA was applied as a foliage application at a rate of 8 g GA/ha as ProGibb® SG liquid fertiliser (40% GA). The GA was diluted with 200 L/ha water along with a surfactant, (Contact™ 0.25 ml/L) and sprayed across the treatment area. The area was left to regrow for 32 days until it reached a minimum pre-grazing compressed height of 9±0.5cm, or 20 clicks of the rising plate meter (RPM, Jenquip F150 Electronic Pasture Meter) which is equivalent to 3300 kg DM/ha based on the standard equation (kg DM/ha = 140 x RPM clicks + 500).

Thirty-six mixed-age, Friesian x Jersey cows were allocated to one of three treatment groups based on covariate milk solids (MS) production (1.4±0.01 kg MS/cow/day), milk protein (0.6±0.01 kg protein/cow/day), live weight (499±1.7 kg) and days in milk (209±0.5 days) which was measured a week prior to commencing the study. Groups were randomly assigned to one of the three pasture treatments and adapted to experimental conditions over six days from 1 May 2014. Temporary electric fencing was used to achieve a target allocation at 35 kg DM/cow/day above ground level based on pasture mass which was estimated using the RPM and winter equation (kg DM/ha = 140 x compressed height + 500). Cows had *ad lib* access to clean water in portable troughs and were milked twice a day at 0600h and 1500h and received a new allocation of pasture after each morning milking.

Herbage measurements

Throughout the study, pasture mass was measured pre- and post-grazing, by harvesting six quadrats per treatment every other day (n=66 quadrats/treatment). All plant material within 0.2 m^2 quadrats was harvested to soil level, washed and the dry weight recorded after force air oven drying to a constant weight at 60°C. Immediately prior to harvesting each quadrat the compressed height of pasture in the quadrat area was measured with the RPM to confirm the pasture mass calibration and determine apparent DM intake (kgDM/cow/day) using the disappearance method.

Calibration equations derived from regression analysis were:

Control kgDM/ha = 92*RPM (r^2=0.91, n=66),
GA kgDM/ha = 83*RPM (r^2=0.93, n=66)
N fertiliser kgDM/ha = 91*RPM (r^2=0.93, n=66).

Herbage samples for nutritive analysis were collected daily by cutting pasture to the target residual grazing height of 3.5 compressed cm at 10 random locations per treatment. The sample was mixed thoroughly and a sub-sample of approximately 20 g fresh weight (FW) was sorted into ryegrass vegetative, ryegrass reproductive, white clover, weeds and dead material. These were then oven dried at 60°C for 48 hours and dry weights recorded. These data were used for botanical composition analysis.

Species (%) = g DM of species/g DM total in the sample

Another 50g FW sub-sample was immediately frozen at 20°C and later freeze dried and ground through a 1-mm sieve (ZM200 rotor mill, Retsch Inc. Newton Pennsylvania, USA). The ground samples were then analysed using near-infrared spectroscopy (NIRSystems 500, Foss, Maryland, USA) to obtain CP, acid detergent fibre (ADF), neutral detergent fibre (NDF), water-soluble carbohydrate (WSC) and digestibility of the organic matter in the DM (DOMD). DOMD was used to calculate metabolisable energy (ME) content of the samples (Geenty et al. 1992).

Animal measurements

Milk yields for each cow were downloaded daily (Delaval Alpro Herd Management system, Hamilton, New Zealand). Milk subsamples were collected at morning and afternoon milking on days 7, 10 and 12 and used to analyse milk fat and protein content (Foss Fourier Transform infrared analyser) and milk-urea N (MUN). Milk-urea N was determined on skimmed milk after cooling and centrifuging to remove fat and analysed by an automated Modular P analyser (Roche/Hitachi). Faecal and urine samples were collected on days 7 and 10 after morning and afternoon milking. Faecal samples were collected by manually stimulating defecation. Urine samples were collected mid-stream after stimulation by rubbing the vulva by hand and acidified immediately with five drops of sulphuric acid to prevent volatilization. Faecal and urine samples were stored frozen (-20°C). Faecal samples were thawed at 4°C for 24 hours. A thoroughly mixed subsample of about 20 g FW was weighed and then oven dried at 100°C for 48 hours, then re-weighed to determine DM%. Another subsample of about 30 g FW was freeze dried for five days at 0.5 mbar, ground through a 1-mm grinder and analysed for N% using an Elemental Analyser (vario MAX CN, Analysensysteme GmbH. Hanau, Germany). Urine samples were thawed at 4°C and analysed for urea, ammonia, N% (Elemental Analyser) and creatinine (kinetic colorimetric assay, Roche Creatinine Jaffe kit).

Statistical analysis

Calibration coefficients for the RPM were derived by linear regression of compressed height and herbage mass. Herbage parameters were analysed using the general-linear-model procedure of Genstat (19^{th} Edition, VSN International Ltd), using sampling date as replicate (since each sampling date was a different allocation) and fertiliser treatment as fixed terms in the model. Milk yield and milk composition parameters from the three herd test days (7, 10 and 12) were analysed for variance by repeated-measures analysis in Genstat using baseline values as covariates. Again, fertiliser treatment was used as the fixed term in the model and animal was the random term. An effect was considered statistically significant if the P value was less than 0.05.

Results

Climate

The cumulative rainfall during the regrowth period, to the grazing period was 142 mm. During the grazing period the cumulative rainfall was 26 mm (Table 1). During the regrowth period the average daily air temperature was

Table 1 Mean (± standard deviation, SD) weekly rainfall (mm), air temperature, and soil temperature to 10 cm below ground level (°C) from the start of the regrowth period (March 27 2014) to the last day of the trial (May 12 2014).

	Temperature				
Week commencing	Soil	SD	Air	SD	Rainfall
27/03/2014	13.2	1.72	14.6	1.55	0
3/04/2014	14.0	1.11	14.0	1.58	49.0
10/04/2014	12.7	1.49	12.3	2.02	15.4
17/04/2014	12.1	1.25	13.2	2.57	40.0
24/04/2014	9.8	1.51	10.5	2.85	56.8
1/05/2014	9.8	1.77	11.8	3.50	0
8/05/2014	8.4	0.92	9.3	0.89	6.0

Table 2 Pre-grazing pasture mass (kg DM/ha), botanical composition (% of biomass) and nutritive value (% of biomass), of untreated pasture (Control) or pasture previously treated with urea (N fertiliser) or gibberellic acid (GA).

Variate	Control	N fertiliser	GA	SEM	P val
Pasture mass	2086	2121	1968	61.6	0.20
Ryegrass	87.8	90.4	85.8	1.88	0.24
Clover	3.31	2.94	4.65	1.11	0.53
Dead	3.47	3.31	4.64	0.91	0.54
Weed	5.44	3.33	4.95	1.50	0.59
Sheath:leaf	20.8	20.3	19.5	1.92	0.89
DM	12.7	11.1	10.4	1.03	0.32
ADF	22.4	22.6	23.0	0.25	0.28
NDF	40.3	39.8	40.7	0.94	0.79
WSC	16.5	15.2	15.6	0.52	0.22
CP	18.8	20.3	19.2	0.67	0.26
WSC:CP	0.88	0.77	0.82	0.04	0.23
DOMD	80.0	79.9	78.5	0.77	0.32
ME (MJ/kg DM)	12.2	12.1	11.9	0.11	0.32

SEM = standard error of the mean, ADF = acid detergent fibre, WSC = water-soluble carbohydrate, NDF = neutral detergent fibre, CP = crude protein, ME = metabolisable energy, DM = dry matter, DOMD = digestible organic matter in the dry matter.

13.2°C with a max. of 17.0°C and a min. of 8.1°C. During the grazing period, the average daily air temperature was 10.5°C with a max. of 17.7°C and a min. of 7.5°C. During the regrowth period the average daily soil temperature to 10 cm at 9am was 12.6°C with a max. of 14.9°C and min. of 8.1°C. During the grazing period, the average daily soil temperature to 10 cm was 9.5°C with a max. of 11.9 and min. of 7.6°C.

Pastures

Due to the low calibration coefficients for the RPM, both the average pre- and post-grazing pasture mass were low, though there was no difference in herbage mass among treatments (Table 2). Similarly, pasture treatment had no effect on botanical composition whereby clover and dead material combined represented less than 10% of the biomass (Table 2). The lack of effect of treatments on herbage mass and botanical composition resulted in no difference in the nutritive value of the pasture. Average DM was low (11.4±1.14%), which influenced the pre-grazing pasture mass. Crude protein content was normal and ranged between 16 and 20% of the DM. Digestibility of organic matter in the DM was 80% resulting in ME of 12.1±0.09 MJME/kg DM.

Intake and milk production

At similar herbage allocation, GA or N fertiliser application in autumn had no effect on DM intake or milk production (Table 3). The daily pasture allocation, above ground level, was 20±1.7 kg DM/cow/day with 72% utilisation, resulting in a mean apparent intake of 14±1.1 kg DM/cow/day. Although there were no differences in milk yield (14.6±0.40 L/day, P=0.70) or MS yield (1.4±0.09 kg MS/cow/d, P=0.13) among treatments, there was a tendency (P=0.06) for greater milk protein content from cows in the control group from those in the GA or N fertiliser treatments. There was no effect of treatment on live weight or BCS.

Nitrogen intake and excretion in faeces, urine and milk

Nitrogen intake (453 g N/cow/day, P=0.75) and faecal DM content (9.1 ± 0.53%DM, P=0.16) were similar among treatment groups (Table 4). Cows grazing pasture treated with GA had higher (P<0.001) faecal-N content (3.72% N) than did the control cows (3.38% N). Urinary-N content (0.4±0.02% N, P=0.33) was similar among treatment groups. Treatments groups had similar ammonia, creatinine, and urea concentrations in urine. Milk-urea N (88.9±1.04 mmol/L, P=0.97) and milk-N excretion (101±5.7 g N/

Table 3 Allocation, dry-matter (DM) intake and milk production of cows grazing untreated pasture (Control), or pasture previously treated with urea (N fertiliser) or gibberellic acid (GA).

Variate	Control	N fertiliser	GA	SEM	P val
Allocation above ground (kg DM/cow/d)	21.9	19.9	18.6	1.85	0.45
Compressed post-graze height (cm)	3.6	3.4	3.7	0.18	0.64
Post graze pasture mass (kg DM/ha)	655	624	611	31.4	0.60
Intake (kg DM/cow/d)	15.0	12.9	13.3	1.04	0.34
ME intake	176	150	154	12.2	0.30
Milk					
Yield (kg)	15.0	14.2	14.6	0.68	0.71
Fat (%)	5.66	5.49	5.00	0.24	0.16
Fat (kg)	0.84	0.77	0.73	0.04	0.17
Protein (%)	4.62	4.39	4.21	0.12	0.06
Protein (kg)	0.69	0.62	0.62	0.03	0.12
Milk solids (kg)	1.53	1.38	1.35	0.07	0.14

SEM = standard error of the mean

Table 4 Nitrogen (N) intake, faecal dry-matter (DM) and N (%), urine ammonia, creatinine, urea and N concentration, milk-urea N (MUN), total N excretion and N use efficiency for milk (NUE) of cows grazing untreated pasture (Control) or pasture previously treated with urea (N fertiliser) or gibberellic acid (GA).

Variate	Control	N fertiliser	GA	SEM	P val
Estimated N intake (g N/c/d)	470	429	460	39.6	0.75
Faeces					
DM %	9.17	9.56	8.51	0.387	0.16
N %	3.38	3.45	3.72	0.066	<0.001
Urine					
Ammonia (mmol/l)	0.94	0.83	0.78	0.077	0.31
Creatinine (mmol/l)	1.39	1.34	1.42	0.096	0.81
Urea (mmol/l)	100.7	91.6	103.2	5.37	0.28
N concentration (g/dL)	4.04	3.73	4.10	0.24	0.33
Milk					
MUN (mmol/L)	88.1	90.1	88.6	4.89	0.95
N excretion (g N/c/d)	108	97.0	96.9	4.40	0.12
NUE for milk (%)	23.0	21.5	24.8	2.00	0.51

SEM = standard error of the mean

cow/d, P=0.12) were similar among treatment groups as was N-use efficiency for milk (23.6 ± 0.01 %, P=0.51).

Discussion

Pasture response to N or GA

There was no response in DM production to the application of N fertiliser or GA in late March through May. The lack of pasture response to either GA or N was an unexpected outcome as climate conditions were suitable for growth. Although the pre-graze herbage mass in May was low, at 2000 kg DM/ha, accumulation from a starting cover of around 690 kg DM/ha after mowing in March indicates a reasonable growth rate of 41 kg DM/ha/d. There was little dead material at the base of the sward and the absence of clover stolon or large tiller populations contribute to the low pasture mass. While this explains the low pasture mass it doesn't explain the lack of treatment effect. Factors that could have contributed to lack of response to N fertiliser and GA include: declining temperatures and day length, high soil N availability from effluent and, with respect to GA, the long regrowth interval. Although the mode of action of N fertiliser and GA differ when applied to pasture, climatic conditions will influence ryegrass growth irrespective of treatment. From application to the start of the grazing study 32 days later, soil temperatures declined from 12.6 to 9.5°C, although still above the threshold for ryegrass growth and day length from 12 to 10 hours. While it is possible to get a response to N fertiliser in autumn there is often a lower response efficiency in autumn than in spring (Cameron et al. 2005; Sun et al. 2008). Of the factors contributing to low autumn response to N, in this study it is likely that N wasn't limiting, the pasture herbage N content was >3% of the DM and the lack of clover in this instance is more likely to reflect increased competition from ryegrass under high soil mineral N from historic effluent application.

Because pasture growth rates decline naturally as daylength and temperatures decline, the response to application of GA in autumn was expected to be less than in spring, based on previous studies (van Rossum et al. 2013; Zaman et al. 2014). Nevertheless, a response was expected, though it is likely that the duration between applying GA and harvesting pasture may be an important determinant in the size of the response relative to the control. Both the observations of Ball et al. (2012) who showed a shift in partitioning from above to below ground and the yield reductions in subsequent regrowth after application reviewed by Matthew et al. (2009) imply that applying exogenous GA has a carbon cost which balances out over time. This study was conducted five to six weeks after the application of GA and, therefore, the initial response to GA may have disappeared over the extended regrowth period.

Milk production and N-use efficiency

There were limited effects of the treatments on milk yield, milk composition or N-use efficiency for milk. Although allocation was lower than target, due to the difference in plate meter calibration for pasture mass (manufacturer versus experimental) used during and after the trial, there was no difference in allocation (20 kg DM/cow/day) or apparent intake (14 kg DM/cow/day) among treatments. With similarities in botanical composition and nutritive value, the diets consumed by each treatment group were very similar. It was expected that diet changes may have affected herbage protein and N-use efficiency for milk, including changes in milk urea concentration as an indication of protein nutrition. The elevated spot faecal N% of cows in the GA group is surprising given the lack of statistical difference in N intake and milk N. From a numerical N-balance perspective, the GA cows had the most surplus N and, perhaps this is why faecal N is higher, although we question the reliability of spot samples in the absence of consistence in other variables such as milk urea, N intake and milk N.

To date, there are no published studies on the effect of GA on animal production. In a previous study (Allen 2010) we treated perennial-ryegrass and white-clover pastures with GA (8g GA/ha) or N fertiliser (50kg N/ha) in early March and, after 29 days regrowth, compared milk yield of cows grazing treated pastures. In that study, there was no effect of GA or N on pasture response or milk yield. However, applying GA in that study reduced herbage N content, increased clover content, and reduced apparent N intake, which has implications on lower urinary N excretion. A key finding of that study and the current study is that replacing N fertiliser with GA in autumn is likely to lead to similar pasture production and maintain milk yield. If soil N is not limiting pasture production, then there is little justification for late season use of N fertiliser. Provided the timing of grazing following GA application is early enough to capture the benefits then there may be value in GA use in the autumn to replace N fertiliser.

Conclusion

The timing of fertiliser application and grazing following application are important factors to consider in farm-management practices. When pasture growth rates are low due to rainfall, temperature, or daylength, it is likely that there will be a limited response in DM production to the application of GA in mid-to-late autumn. Minimal changes to DM production and botanical composition resulted in no difference in nutritive value and, in this study animal performance was not affected by the application of GA in late autumn.

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