Response of simple grass-white clover and multi-species pastures to gibberellic acid or nitrogen fertiliser in autumn

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Abstract

Herbage dry matter (DM) production, botanical composition and nutritive value of pastures were measured in response to the application of 24 g/ha gibberellic acid (GA; 60 g ProGibb®/ha) and 50 kg N/ha nitrogen fertiliser (N; 106 kg urea/ha) in autumn. Responses were compared for simple two species grass (perennial ryegrass or tall fescue)-white clover pastures and multi-species pastures where herbs (chicory and plantain), legumes (red clover and lucerne) and prairie grass were sown with the simple mixtures. Four weeks after application, the increase in DM yield averaged across pasture mixtures relative to untreated plots was 273, 104, and 493 kg DM/ha for GA alone, N fertiliser alone, and GA and N combined, respectively. Application of GA increased the white clover percentage in all pastures except simple tall fescue pastures. The percentage of herbs in multi-species pastures was increased by application of N but not GA. Crude protein concentration was decreased by GA application in all pastures except simple ryegrass pastures. The results from this study show that it is important to consider the botanical composition of pastures when determining the effect of GA on DM yield and nutritive value.

Keywords: crude protein, *Cichorium intybus*, *Festuca arundinacea,* gibberellins, *Lolium perenne*, *Plantago lanceolata*, nutritive value

Introduction

Pastoral farming in New Zealand is driven by seasonal feed supply and demand, and the amount of pasture that is grown on farm. In dairy herds, cows are required to sustain a high level of milk production over a long lactation. Lactation length is influenced by feed supply and extra days in milk in the autumn increases total milk yield. To increase pasture production in autumn, farmers often apply nitrogen (N), but this can have adverse environmental impacts due to losses of N from urine via leaching from pasture (Di & Cameron 2002). The problem is exacerbated as cooler temperatures reduce plant growth and N concentration of herbage accumulates. As animal demand for protein declines, the proportion of N excreted in urine increases, and urine deposited in autumn is subject to greater leaching losses due to high winter drainage (De Klein 2001; Di & Cameron 2002).

Increasing public and government pressure to reduce N leaching has prompted investigations into alternative fertilisers and pasture species which can lead to increased dry matter production of pastures with reduced herbage N concentrations. Recent comparisons of simple perennial ryegrass-white clover and tall-fescue-white clover pastures with multi-species pastures (MSP) containing additional herbs and legumes, have shown greater annual dry matter (DM) yield (Nobilly *et al*. 2013) and reduced urine N excretion from dairy cows (Totty *et al*. 2013) with MSP. The plant growth hormone, gibberellic acid (GA), may also present an opportunity to increase pasture production when pasture growth is limited (Matthew *et al*. 2009) and could be used as an alternative to N fertilisers in spring and autumn. A review of plant growth responses to GA showed that the median response to GA was 500 kg DM/ha when applied once with rates between 25 and 700 g active ingredient/ha (Matthew *et al*. 2009). However, there is limited published information (Morgan & Mees 1958; Scott 1959) available on the effects of GA applied at low rates on pasture production in autumn, and none on how it might affect DM yield response and botanical composition of multi-species pastures. The objectives of this study were to compare the effects of GA and N application on DM production and nutritive value of simple and MSP in autumn.

Materials and Methods

Experimental site and design

The experiment took place on the Lincoln University Research Dairy farm in Canterbury, New Zealand (43°38´S, 172°27´E) between 23 March and 26 April 2011. The soil at the site is a Paparua sandy loam. Soil tests taken before the experiment began showed: pH = 5.8, Olsen P = 33 mg/L, K = 0.38 me/100 g, Ca = 6.4 me/100 g, Mg = 0.46 me/100 g and S = 0.16 me/100 g. The pasture was sown in February 2010 and at the time of the experiment was 13 months old. The experimental area was under full irrigation and had previously been rotationally grazed by dairy cows. The experiment was a split plot factorial design laid out in three randomised blocks. The main plots were factorial combinations of two base grasses (ryegrass (RG) or tall fescue (TF)) and two levels of diversity (simple (base pasture plus clover), and base pasture plus herbs and legumes (MSP). Table 1 shows the plant species and cultivars in the main plot treatments. The four sub-plot factor combinations were: control (CNT, no GA or N); GA (24 g GA/ha); N (50 kg N/ha); and GA + N (24 g GA/ha + 50 kg N/ha). Nitrogen was applied in the form of urea (47% N) and granules were applied by hand. Gibberellic acid was applied by spray applicator using ProGibb-SG^{TM} (40% GA) and a surfactant (Contact^{TM}), both from NuFarm Ltd.

Gibberellic acid was applied at a higher rate than the manufacturers recommendation of 8 g GA/ha as a result of slower application speed than that used during sprayer calibration. The higher rate of 24 g GA/ha was consistent for all plots.

Experimental areas were situated within a larger grazing trial consisting of three 0.5 ha paddocks of each of the four pasture treatments (see Nobilly *et al.* (2013) for site description). Electric polywire was used to separate a 10 ×10 m experimental area within each 0.5 ha paddock. On 23 March 2011, the experimental area was mown to 4 cm stubble height and the four sub-plot areas of 2 × 8 m were marked out with wooden pegs. Fertiliser treatments were randomly allocated to sub-plots and were applied within 48 hours of mowing. Plots were allowed to regrow until final harvest four weeks later on 26 April 2011.

Pasture measurements

Dry matter yield at the end of the 4 week regrowth period was determined from one 0.4 × 8 m cut taken with a rotary mower to 4 cm above ground level. The fresh weight (FW) of herbage was recorded and a subsample of approximately 100 g FW from each plot was taken to determine DM%.

Snip samples of vegetation were taken from each plot at 4 cm height and two sub-samples (200g FW) were removed. One sample was freeze dried and ground through a 1 mm sieve prior to analysis using near infrared spectroscopyto measure the concentration of crude protein (CP), organic matter (OM), water-soluble carbohydrate (WSC), acid detergent fibre (ADF) and neutral detergent fibre (NDF) by the Lincoln University Analytical Laboratory. Metabolisable energy content was calculated using the modified ADF equation for forages (CSIRO 2007). A second sample was sorted into species and dead material, oven-dried at 65°C, and percentage botanical composition on a DM basis was determined.

Statistical analysis

In week three of the regrowth period, cows broke the fence and grazed three of the four plots in one of the simple RG blocks. The three affected plots were treated as missing values in the analysis. Herbage DM yield, nutritive value and botanical composition at final harvest were analysed using ANOVA of a split plot design using Genstat (v. 12.2). Main plot factors were base grass × diversity and sub-plot factors were the GA × N applications. Analysis of botanical composition of herbs, legumes other than white clover and prairie grass were performed for MSP treatments only.

Results

Rainfall and temperature

The maximum temperatures for the experimental period ranged from 10.4 to 24.8°C (mean 16.9°C) and minimum temperatures ranged between 3.4 and 13.2°C (mean 7.8°C). Mean soil temperature at 10 cm was 11.1°C. Over the regrowth period the accumulated rainfall was 100 mm (no irrigation applied). Day length reduced from 12.1 hours on 23 March to 10.3 hours on 26 April.

Dry matter yield and botanical composition

The mean DM yield across all treatments was 1230 kg DM/ha (Table 2). There were no interactions between GA × N or between base grass × N or base grass × a).

Table 1 Pasture species and cultivars in the four pasture mixtures used in the experiment.

Species diversity	Base Grass	
	Perennial ryegrass	**Tall Fescue**
Simple	Diploid perennial ryegrass (cv. 'One50') Large Leaf White clover (cv. 'Kopu II')	Summer active tall fescue ('Advance') Large Leaf White clover (cv. 'Kopu II')
Multi-species	Diploid perennial ryegrass (cv. 'One50') Large Leaf White clover (cv. 'Kopu II') Prairie grass (cv. 'Atom') Chicory (cv. 'Choice') Plantain (cv. 'Tonic') Red Clover (cv. 'Colenso')	Summer active tall fescue ('Advance') Large Leaf White clover (cv. 'Kopu II')Prairie grass (cv. 'Atom') Chicory (cv. 'Choice') Plantain (cv. 'Tonic') Lucerne (cv. 'Torlesse')

Compared with non-GA treated pasture, the application of GA increased DM yield by 31% (1060 vs 1393 ± 54 kg DM/ha, P<0.05). Application of N fertiliser also increased DM yield by 14% (1308 vs 1146 ± 54 kg DM/ha, P = 0.05) which was less pronounced compared with GA.

The base grass species were more abundant in simple than MSP (79% vs 28%, P<0.001). Perennial ryegrass was a more (P<0.05) abundant base pasture than tall fescue, accounting for 60% of the DM across simple and MSP compared with 45% for tall fescue. The proportion of clover (10% vs 4% white clover) and herb (43% vs 27% herb) was higher in TF-based than RG-based pastures.

In TF based simple pastures, application of GA resulted in a decrease in the proportion of white clover, whereas in all other treatments the proportion of white clover was increased by GA application (base grass × diversity × GA interaction, P<0.05, Table 2). In both RG and TF-based pastures, the proportion of herbs increased from 31.8% to 37.9% of the DM when N fertiliser was applied (P<0.05). A base grass × GA interaction (P = 0.013) showed that the proportion of herbs increased with GA in RG-based swards (23 to 31%) but reduced with GA in TF-based swards (47% to 39%).

Table 2 Effect of gibberellin (GA), nitrogen (N) or no treatment (CNT), on mown pasture yield (kg DM/ha) and botanical composition (% total DM) of simple and multi-species pastures sown with a base grass of ryegrass or tall fescue. SEM = standard error of the mean is for the B x D x GA interaction. P values from ANOVA are shown at the base of table.

Base pasture and diversity	Fertiliser	Botanical Composition						
		Yield	Base grass	White clover	Dead	Other legume	herb	Prairie grass
Simple								
Ryegrass	CNT	1185	86.3	0.6	12.7	-	-	-
	GA	1650	74.3	17.9	7.7	-	-	-
	N	1583	76	1.3	18.1	-	-	-
	GA + N	1851	86.2	3.5	10.2	-	-	-
Tall fescue	CNT	1089	72.1	21.8	6	-	-	-
	GA	1338	78.9	13.3	5.3	-	-	-
	N	1028	76	15.4	8.5	-	-	-
	GA + N	1672	86.4	9.6	3.8	-	-	-
Multi-species								
Ryegrass	CNT	960	51.6	0.7	8.3	5.8	21.8	11.8
	GA	1108	31.8	6.3	4.2	13.3	29.3	15.2
	N	1004	50.2	1.9	5.8	8.3	24.1	9.8
	GA + N	1280	42.5	2.7	5.2	6.4	32.5	10.7
Tall fescue	CNT	804	15	3.2	8.6	6.2	45.7	21.3
	GA	1036	13.5	4.1	9.3	6.9	30.4	35.5
	N	840	10.8	6.7	5.5	8.1	48.1	20.6
	GA + N	1208	8.5	9.3	3.2	8.9	46.9	23.3
	SEM	141.6	4.67	2.38	4.0	3.65	3.43	7.83
P value								
	Base (B)	0.23	0.012	0.008	0.98	0.8	<.001	0.26
	Diversity (D)	0.04	<.001	0.008	0.97	NA*	NA	NA
	B x D	0.60	0.03	0.09	0.8	NA	NA	NA
	GA	<.001	0.5	0.29	0.004	0.45	0.95	0.22
	N	0.05	0.6	0.22	0.59	0.96	0.05	0.26
	D x GA	0.34	0.02	0.74	0.07	NA	NA	NA
	D x N	0.47	0.6	0.04	0.01	NA	NA	NA
	B x D x GA	0.98	0.71	0.04	0.25	NA	NA	NA

*NA, not assessed as not present in mixture

Nutritive value

Multi-species pastures had lower OM (864 vs 884 ± 3.7 g/kg DM; P<0.01) and lower crude fibre (305 vs 403 ± 11.2 g NDF/kg DM. P<0.001) concentration than simple pastures (Table 3). TF-based pastures had a lower concentration of ADF (206 vs 227 ± 3.4 g ADF/kg DM. P<0.01) and higher ME (11.4 vs 11.0 ± 0.05 MJ ME/kg DM. P<0.01) than RG-based pastures.

Crude protein (CP) concentration ranged between 157 and 233 g/kg DM across treatments but there was no effect of base grass or diversity on crude protein concentration (193 g/kg DM, Table 3). Nitrogen fertiliser increased mean CP concentration from 182 to 198 ± 3.0 g/kg DM (P<0.01). A three way interaction between base grass × diversity × GA for CP concentration (P<0.01) revealed that the application of GA resulted in a 15 g CP/kg DM reduction in MSP compared with a 10 g CP/kg DM increase in simple pastures. However, in simple pastures the positive CP response was more pronounced in RG than TF based pastures. The variation in CP concentration between simple and MSP was compensated for by increased (10.2 g WSC) WSC concentration in MSP and reductions (9.2 g WSC) in simple pastures.

Table 3 Effect of gibberellin (GA), nitrogen (N) or no treatment (CNT), on chemical composition (g/kg DM) and metabolisable energy content (MJ ME/kg DM) of simple and multi-species pastures sown with a base grass of ryegrass or tall fescue. SEM = Standard error of mean is for the B x D x GA interaction. P values from ANOVA are shown at the base of table.

Base pasture and diversity	Fertiliser	Chemical composition					
		OM	NDF	ADF	CP	WSC	ME
Simple							
Ryegrass	CNT	894	458	253	133	176	10.7
	GA	888	404	227	185	161	11.1
	N	881	432	241	165	146	10.8
	GA + N	880	425	239	174	150	10.9
Tall Fescue	CNT	884	368	200	218	146	11.5
	GA	882	407	211	212	139	11.3
	N	882	367	201	233	146	11.4
	GA + N	882	388	206	223	141	11.4
Multi-species							
Ryegrass	CNT	876	386	237	150	167	10.9
	GA	876	359	234	160	158	10.9
	N	863	296	203	205	142	11.4
	GA + N	880	306	212	185	159	11.0
Tall Fescue	CNT	863	299	211	180	138	11.3
	GA	869	317	215	186	138	11.2
	N	861	279	195	219	133	11.5
	GA + N	872	258	209	185	158	11.3
	SEM	7.26	17.0	57.7	10.2	7.14	0.12
P values							
	Base (B)	0.75	0.06	0.004	0.07	0.42	0.001
	Diversity (D)	0.009	<0.001	0.11	0.21	0.89	0.27
	B x D	0.51	0.85	0.04	0.40	0.75	0.10
	GA	0.58	0.61	0.38	0.77	0.9	0.36
	N	0.5	0.07	0.06	0.002	0.38	0.04
	D x GA	0.28	0.44	0.1	<0.001	0.02	0.08
	D x N	0.77	0.13	0.07	0.05	0.51	0.05
	B x D x GA	0.85	0.03	0.12	0.005	0.86	0.1

OM, organic matter; NDF, neutral detergent fibre; ADF, acid detergent fibre; WSC, water-soluble carbohydrate; CP, crude protein; ME, metabolisable energy

Discussion

Dry matter yield

Compared with non-GA treated pastures, GA application increased DM yield in autumn by 331 kg DM/ha (31%) when measured 4 weeks after application. These results are in line with previous studies regarding increased DM yield following GA application in autumn (Morgan & Mees 1958; Scott 1959; Edmeades & McBride 2012). Across seasons, a wide range of DM yield increases to GA application have been reported ranging from 50 to 980 kg DM/ha for temperate grasses (Wittwer & Bukovac 1957; Morgan & Mees 1958; Finn & Nielsen 1959; Scott 1959; Blacklow & McGuire 1971; Matthew *et al*. 2009; McGrath & Murphy 1976). Previous studies have demonstrated reductions in DM yield at successive harvests following GA application (Morgan & Mees 1958; Finn & Nielsen 1959) which is often associated with reduced tiller densities. However, no attempt was made to determine tillering behaviour or subsequent regrowth of treatments in the current study.

The results from the current study also showed increased DM yield after N fertiliser application, but the response was less pronounced (14% increase) than from GA application. Previous studies have noted that relative responses to GA are greater at lower temperatures (Arnold *et al.* 1967; Blacklow & McGuire. 1971) and at higher N fertility (Ball *et al*. 2012). This was confirmed in the current study, where GA applied with N achieved the maximum DM yield.

Botanical composition

The effect of GA on botanical composition was dependent on the sensitivity of the base grass to GA application relative to either herbs or legumes. In this study, base grasses were responsive to GA in simple pastures (base grass increased by 450 ± 121 kg DM/ha) but not in MSP (base grass decreased by 36 ± 59 kg DM/ha). A range of grass species have been shown to respond positively to GA (Wittwer & Bukovac 1957; Finn & Neilson 1959; Laude *et al*. 1960), although the relative response tends to be greater in tall fescue than ryegrass (Wittwer & Bukovac 1957). This was demonstrated in the current study by the clover response in RG and TF simple pastures. A strong response to GA by tall fescue resulted in a reduction in white clover content in TF pastures while a moderate response to GA by ryegrass resulted in an increase in white clover in RG pastures. In a comparison of grasses and legumes responding to different GA rates, Finn & Nielson (1959) showed that legumes were more responsive to GA than grasses, particularly at very high rates of GA (295 to 590 g/ha). On the other hand, herbs did not respond to GA (herbs increased by 66 ± 77 kg DM/ha) but accounted for a higher proportion in the pasture when N was applied (herbs increased by 180 ± 74 kg DM/ha). Chicory and plantain showed a positive response to GA application in previous studies (Wittwer & Bukovac 1957; Dijkstra *et al*. 1990) but these results were in spring when the plant became reproductive. The evidence from this and previous studies show that botanical changes and yield of pastures after GA application will be influenced by the abundance of the most responsive plant species when GA is applied. The size of the response then is subject to season (particularly in the case of herbs), N fertility and climatic conditions.

Nutritive value

Metabolisable energy was higher in TF based than RG based pastures (11.3 vs. 10.9 ± 0.05 MJME/kg DM), which may be accounted for by the high content of clover. There was little effect of GA on ME; this reflects that GA application did not alter ADF concentration from which ME was calculated (CSIRO 2007).

The effect of GA on CP concentration of simple and MSP also appeared to be linked to changes in the clover content of the pastures. In MSP, where the clover content was low (<10%) and increased only marginally due to GA application, the concentration of CP was reduced with GA application irrespective of the base grass. In simple pastures, there was a reduction in the CP concentration of TF based pastures because white clover content decreased; in contrast, in simple RG based pastures, white clover content increased from 1% to 11% due to GA, therefore increasing overall crude protein concentration. Reductions in crude protein following GA application have been shown in some cases (Finn & Nielsen 1959; Brown *et al*. 1963), but not all (Morgan & Mees 1958; Matthew *et al*. 2009; Parsons *et al*. 2013). In MSP it is difficult to attribute change in the CP to one particular species but increased overall DM yield without appreciable change in legume content has in this case reduced CP concentration.

Summary

A single GA application to conventional grass and clover pastures offers an opportunity to improve DM production in autumn. Responses of pastures containing a high proportion of herbs may not achieve the same yield benefits as grass/clover pastures. However, from an environmental standpoint, autumn is a critical period in which N leaching losses from urine patches are high and amenable to manipulation by reducing the CP concentration of the diet. Provided there is sufficient CP in the diet to meet animal requirements the reduction in CP concentration in both simple and multi-species pastures following GA application offers an opportunity to reduce N losses.

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