Seed yield of three perennial ryegrass cultivars following treatment with Moddus[®] straw shortener

R.J. Chynoweth¹ and D.J. Moot²

¹Foundation for Arable Research, PO Box 23133, Templeton 8445, New Zealand ²Faculty of Agricultural Sciences, Lincoln University, PO Box 89084, Canterbury, New Zealand

Abstract

First year crops of three diploid perennial ryegrass (Lolium perenne) cultivars, 'Meridian', 'Bronsyn' and 'Grasslands Impact', that contained the AR1 endophyte, were sown on 1 April and 14 May 2008. Moddus[®] (a.i. 250 g/l trinexapac-ethyl) plant growth regulator at 0, 800 and 1600 ml/ha was applied at Zadoks growth stage 32. Moddus[®] increased seed yield by approximately 26% for each 800 ml/ha increment applied from 1715 (0 ml/ha) to 2195 (800 ml/ha) and 2720 kg/ha (1600 ml/ha). The seed yield increase was achieved through an increased number of seeds/ m^2 . Total stem length was reduced from 0.75 m to 0.66 m by application of 800 ml/ha of Moddus[®] and further to 0.60 m with 1600 ml/ha. Moddus[®] decreased absolute lodging at harvest and increased the harvest index from 12.0 to 17.6%, primarily through an increase in seed mass/ha. Straw mass remained constant for 'Meridian' and 'Bronsyn' but in 'Grasslands Impact' the increase in harvest index was limited through an associated increase in straw DM with each Moddus® application. These results suggest the highest application rate of 1600 ml/ha should be used to increase seed yield of perennial ryegrass, and further research should examine the impact of even higher Moddus[®] rates.

Additional keywords: harvest index, trinexapac-ethyl, Lolium perenne, sowing date

Introduction

Perennial (*Lolium perenne* L.) and hybrid ryegrass (*Lolium x hybridum* Hausskn) seed crops are an important component in the crop rotation of New Zealand arable farmers. They provide a modest return per hectare (ha), through grazing or silage in their vegetative state, but seed production provides approximately 85% of the total economic return (Rolston and Archie, 2005). Plant growth regulators (PGRs) have been the focus of investigations in perennial ryegrass seed crops for many years (Hebblethwaite *et al.*, 1980; Hampton *et al.*, 1987; Chastain *et al.*, 2003; Rolston *et al.*, 2004). Between 1980 and 1987 work focused on the use of paclobutrazol which showed increases in seed yield of between 8 and 136% (Wiltshire *et al.*, 1987). However the chemical properties of paclobutrazol subsequently resulted in possible yield reductions in some following crops and therefore its use is not widespread. The introduction of Moddus[®] (trinexapac-ethyl), a foliar absorbed PGR, revolutionised grass seed yields in New Zealand and it is currently used by >95% of seed growers. Seed yield increases of 50% are common in New Zealand (Rolston *et al.*, 2004; Chynoweth *et al.*, 2010) and Oregon, USA (Chastain *et al.*, 2003) although application rates in the USA are often lower than those applied in New Zealand. Plant growth regulators are applied with the goal of increasing seed yield through a reduction in stem length which reduces lodging and allows greater seed set (pollination) and more efficient use of solar radiation (Hebblethwaite, 1980; Griffith, 2000). Hebblethwaite *et al.* (1978) reported a 61% yield increase using wires that physically prevented lodging, but PGRs provide a more practical option.

Cultivars were selected to be diploid, contain the same endophyte (AR1) and to provide a 35 day range in expected head emergence date. 'Meridian', a cross between 'Kangaroo Valley', an Australian ecotype, and 'Yatsyn 1' reaches head emergence approximately 17 days earlier than 'standard' cultivars and was first entered into the Seed Certification Scheme in 1998. 'Bronsyn' reaches head emergence at the 'standard' time and is a selection from a 'Mangere pasture'. 'Bronsyn' was first entered into the Seed Certification Scheme in 1997. 'Grasslands Impact' reaches head emergence approximately 21 days later than 'standard' cultivars. 'Grasslands Impact' was selected from crossing late heading plants of 'Grasslands Nui' with an ecotype imported from North West Spain. 'Grasslands Impact' was breed as a true perennial, but was classified as a 'long rotation hybrid' due to a low percentage of seed containing tip awns when first entered into the Seed Certification Scheme in 1992.

This experiment was designed to determine the response of three ryegrass cultivars which mature under different environmental conditions, to the application of three rates of Moddus[®].

Materials and Methods

The experiment was located in paddock 9 of Iversen field at Lincoln University, Canterbury, New Zealand (43° 38' 51" S, 172° 28' 00" E), on a Wakanui silt loam soil (Cox, 1978). The previous cropping history was spring barley (*Hordeum vulgare* L.), preceded by four years of lucerne (*Medicago sativa* L.).

Individual plots were 10 x 2.1 m arranged in a split-split plot design with three replicates. The main plot treatment was sowing date (1 April and 14 May 2008), sub plot treatment was cultivar ('Meridian', 'Bronsyn' and 'Grasslands Impact') and the sub-sub plot treatment was application rate of Moddus[®] at 0, 800 or 1600 ml/ha. Sowing rate was calculated to obtain a target plant population of 300 plants/m².

Weed control was achieved by the use of $1.5 \text{ l/ha Nortron}^{\textcircled{8}}$ (a.i. 500 g/l ethofumesate) applied pre emergence followed by 1.5 l/ha Jaguar⁸ (a.i. 25 g/l diflufenican and 250 g/l bromoxynil) on 20 May 2008. All plots were mechanically defoliated to 60 mm as required to keep vegetative dry matter below approximately 3500 kg/ha (the limit of mechanical defoliation and to surrogate for herbage removal as silage). The final defoliation occurred when approximately 5% of tillers removed had nodes present. This was equivalent to Zadoks growth stage 30 (Zadoks *et al.*, 1974), and was an average of all tillers.

Moddus[®] was applied at Zadoks growth stage 32 (Table 1) in a mix with 200 ml/ha Proline[®] (a.i. 250 g/l prothioconazole) for disease control. The Zadoks growth stage 32 was determined from sequential destructive harvests of 50 tillers per sub plot dissected lengthwise so nodes could be visually assessed. Application was made through a motorised boom sprayer at 200 l of water/ha using 110°, 04 flat fan nozzles at 300 kilopascals (kpa) of pressure.

Nitrogen was applied in three applications based on nodal development with a target N application rate of 180 kg/ha (Table 1). Soil mineral N (ammonium and nitrate) was measured at 21 kg/ha to a 0-300 mm depth on 1 September 2008. The first application of 50 kg N/ha occurred on 10 September 2008 as calcium ammonium nitrate (CAN, 27% N) using a tractor mounted boom spreader. The second N application occurred following the final defoliation and the third N application approximately 21 days later, both as urea (46% N). The three cultivars have different ear emergence dates which lead to variation in nodal development and therefore N application dates (Table 1). The timing of N was aimed to avoid inducing rapid growth during early stem extension which leads to early lodging.

Table 1: Final defoliation date, Moddus[®] application date, and nitrogen application rates and dates for all main plot treatments of three perennial ryegrass cultivars sown on two dates at Lincoln University, Canterbury in the 2008-09 growing season.

dates at Encom University, Canterbury in the 2008-09 growing season.							
Sowing		Final	Moddus [®]	Nitrogen application rate (kg N/ha)			
date	Cultivar	defoliation	application date	and date app	plied in parent	thesis (2008)	
(2008)		date (2008)	(2008)	$1^{st} dose^1$	2 nd dose	3 rd dose	
1/4	'Meridian'	6/9	26/9	50 (10/9)	75 (28/9)	55 (7/10)	
	'Bronsyn'	22/9	15/10	50 (10/9)	75 (7/10)	55 (23/10)	
	'Grasslands	7/10	21/10	50 (10/0)	75 (16/10)	55(14/11)	
	Impact'	//10	21/10	30 (10/9)	75 (10/10)	33 (14/11)	
14/5	'Meridian'	29/9	15/10	50 (10/9)	75 (28/9)	55 (7/10)	
	'Bronsyn'	7/10	23/10	50 (10/9)	75 (16/10)	55 (11/11)	
	'Grasslands	15/10	28/10	50 (10/9)	75 (23/10)	55 (18/11)	
	Impact'	13/10					

¹as calcium ammonium nitrate (27% N, 8% Ca), all other applications as urea (46% N).

Disease control was achieved through applications of Proline[®], Kocide[®] 2000DS (a.i. 350 g/kg copper) and AtomicTM (a.i. 105 g/litre epoxiconazole and 420 g/litre carbendazim) throughout flowering and seed development. Irrigation was applied via a big gun travelling irrigator or a rotating boom travelling irrigator as required ensuring measured soil moisture deficit did not exceed 65 mm. A total of 220 mm was applied between 11 November 2008 and 9 January 2009.

At 40% seed moisture content (SMC) (determined on hand collected samples by oven drying at 130°C for one hour (ISTA, 2009)) three rows by 1 m in length were cut at ground level from two locations within each plot. Within each quadrat two 0.3 m sections of row were removed for harvest component analysis and then returned to the bulk sample for seed yield assessment. Cut samples were naturally dried by hanging over an outdoor drying rack in hessian bags. After approximately 14 days, bags were collected and stored indoors. Three months following harvest, samples were threshed using a Wintersteiger stationary thresher with all seed collected for further processing. Seed was cleaned to a first certification standard generation seed (AsureQuality, 2008) using a screen cleaner for the removal of straw. A Dakota seed blower was then used to sort seed into fractions where individual seed was either lighter or heavier than 1.5 mg. To confirm seed weights, 200 individual seeds from each weight fraction were weighed to four decimal places. Seed greater than 1.5 mg was classed as 'first grade' seed which was considered sufficiently heavy to be saleable. Seed <1.5 mg was re-cleaned to remove empty lemma and palea fractions to achieve an individual seed weight ranging from 0.5-1.5 mg. This was considered 'second grade' seed. All seed yields are reported at 11% SMC while all straw weights are reported as dry weights. Harvest index (HI) was calculated by adjusting seed yields to 0% SMC and straw weights expressed in kg DM/ha.

Seed head numbers were determined by counting the number of heads present in two 0.3 m sections of row from two locations in each plot. The two samples were averaged and multiplied to per m^2 . Stem length, internode length and the number of spikelets/head were measured on 50 stems from each sample (100 stems/plot). Thousand seed weight was determined from 500 seeds from the final machine dressed sample. Lodging was assessed in all plots every 4-6 days. A score of 0% meant the plot was fully vertical, 50% meant the plot on average was leaning on a 45° angle while a 100% score indicates the entire plot was lying horizontal.

All statistical analyses were performed using Genstat[®] (version 14, VSN International Ltd, UK). All plot data were analysed using an analysis of variance (ANOVA) with means separation achieved using the least significant difference (LSD) test.

Results

Total seed yield

The total seed yield (≥ 0.5 mg) was affected (P<0.05) by sowing date and Moddus[®] rate (Table 2) but not their interactions or cultivar. Sowing on 14 May produced 2980 kg/ha of seed compared with 2430 kg/ha from sowing 1 April (11% SMC). The application of Moddus[®] increased (P<0.05) yield from 2125 kg/ha in the untreated control to 2745 kg/ha at 800 ml/ha and 3250 kg/ha at 1600 ml/ha (LSD_{0.05}=190). **Table 2:** Total seed yield (kg/ha) (≥0.5 mg individual seed weight at 11% moisture content) of perennial ryegrass crops sown on two dates and treated with three Moddus[®] rates (a.i. 250 g/l trinexapac-ethyl) at Lincoln University, Canterbury, in the 2008-09 growing season.

Moddus [®] (ml/ha)		(Moddus [®] Moon			
Moduus (IIII/IIa)		1 April		14 May	Wioucus Wiean	
0		1880		2370	2125 с	
800		2420		3070	2745 b	
1600		2995		3500	3250 a	
Sowing date mean		2430 b		2980 a		
Effect	P<		SEM	LSD _{0.05}		
Sowing date	0.05		45.1	274		
Moddus [®]	0.001		65.4	190		

Note: bold figures give the sowing date comparison while means within columns or rows followed by the same letter do not differ (LSD $_{0.05}$).

First grade seed

First grade seed yield (\geq 1.5 mg) was affected (P<0.05) by the main effects of sowing date, cultivar and Moddus[®] rate but none of their interactions. However, all data are presented for clarity (Table 3). On average, 545 kg/ha less seed was produced from sowing on 1 April compared with 14 May. First grade seed yield was higher from 'Bronsyn' than 'Grasslands Impact'. Moddus[®] rate showed a 28% increase in first grade seed between 0 and 800 ml/ha and a further 25% increase at 1600 ml/ha.

The proportion of seed converted into first grade seed was only affected (P<0.05) by cultivar as a main effect. Both 'Meridian' and 'Bronsyn' converted approximately 85% of total seed to first grade seed compared with 73% for 'Grasslands Impact' (LSD_{0.05}=6.2%).

Table 3: First grade seed yield (kg/ha) of perennial ryegrass crops from three cultivars sown on two dates and treated with three Moddus[®] rates (a.i. 250 g/l trinexapac-ethyl) at Lincoln University, Canterbury in the 2008-09 growing season.

				2	\mathcal{O}	\mathcal{O}			
Sowing date	1 April 2008		- Cultivar -	14 May 2008			Cultivar	Cultiva	
Culting	Moddus [®] rate (ml/ha)			Moddus [®] rate (ml/ha)				r mean	
Cultivar	0	800	1600	(Kg/IId)	0	800	1600	(Kg/IIa)	(kg/ha)
'Meridian'	1325	1835	2475	1880	2060	2795	3055	2635	2260 ab
'Bronsyn'	1490	2135	2665	2095	2150	2720	3385	2750	2425 a
'Grasslands	1525	1700	2280	1840	1735	1985	2475	2065	1050 h
Impact'	1555	1700	2280						1930 0
Moddus [®] mean	1450	1890	2470	1940 b	1980	2500	2970	2485 a	
Effect	P<			SEM		LSD _{0.05}			
Sowing date	0.01			17.7		108			
Cultivar	0.05			97.2		317			
Moddus [®] 0.001			67.1		196				

Note: bold figures indicate sowing date comparison.

Lodging

The absolute level of crop lodging (at harvest) was affected by Moddus[®] (P<0.01; F=112) rate and a sowing date by cultivar interaction (P<0.05; F=6.05) but none of the Moddus[®] interactions. The application of Moddus[®] reduced crop lodging from 95% (effectively horizontal) for 0 ml/ha Moddus[®] to 56%

(leaning on a 45° angle) for 800 ml/ha and 35% for 1600 ml/ha (LSD_{0.05}=8.4%). In the sowing date by cultivar interaction (Table 4), 'Bronsyn' and 'Grasslands Impact' sown on 1 April showed a higher (P<0.05) level of lodging compared with sowing on 14 May. Lodging for 'Meridian' was similar (66%) for both sowing dates.

Table 4:Crop lodging (%) at harvest for three perennial ryegrass cultivars sown on two dates
at Lincoln University, Canterbury in the 2008-09 growing season.

•	•				
Sowing date	Cultivar		lodging % ¹		
	'Meridian'	,	67 a		
1 April	'Bronsyn'		66 a		
	'Grassland	ls Impact'	75 a		
	'Meridian'	,	64 a		
14 May	'Bronsyn'		47 b		
	'Grasslands Impact'		53 b		
Effect	P value	SEM	LSD _{0.05}		
Sowing data y cultivar	0.025	2.03	Within cultivar 9.5		
Sowing date x cultival	0.025	2.95	between cultivar 13.0		

 $^{1}50\%$ = crop leaning 45°, 100% = fully horizontal (lodged).

Number of first grade seeds/m²

The number of first grade seeds per m^2 was affected by the main effects of sowing date (P<0.05) and Moddus® (P<0.01) rate but none of the interactions. The application of Moddus® increased the number of first grade seeds from approximately 71,000 seed/ m^2 for the control to approximately 91,600 seed/m² for 800 ml/ha and up to seed/m² 113.600 for 1600 ml/ha (LSD_{0.05}=9,136). Sowing on 14 May (99.660 seeds/ m^2) produced more (LSD_{0.05}=8,260) seeds per m² than sowing on 1 April ($84,480 \text{ seed/m}^2$).

Crop harvest index

Harvest index was influenced by the interaction (P=0.012) between cultivar and Moddus[®] rate (Table 5) and the main effect of sowing date (P<0.01). The HI of both 'Meridian' and 'Bronsyn' increased by

approximately 7% as Moddus[®] rate increased from 0-1600 ml/ha, compared with a 2.2% increase for 'Grasslands Impact'. The HI increased as Moddus[®] rate increased from both sowing dates by approximately 7% between the 0 and 1600 ml/ha Moddus[®] treatments. Crops sown on 14 May had a higher HI (16.9%) than those sown on 1 April (13.0%). The late flowering cultivar, 'Grasslands Impact' had a lower HI than the two earlier flowering cultivars.

Stem length

Moddus[®] was the only treatment to influence total stem length which reduced (P<0.001) from 0.75 m to 0.66 m with 800 and to 0.60 m with 1600 ml/ha (LSD_{0.05}=0.036). On average, 98 mm of height was removed per 1000 ml/ha of Moddus[®] applied.

Season at Line	com oniversity, cunter	oury.			
Cultivor		Cultivar			
Cultival	0	800	1600	mean	
'Meridian'	12.3	15.5	18.9	15.4 a	
'Bronsyn'	12.8	17.1	20.1	15.5 a	
'Grasslands Impact'	10.9	13.1	13.2	12.4 b	
Moddus [®] mean	12.0	15.2	17.6		
Effect	P value SEM	LSD _{0.05}			
Cultiver y Moddue [®]	0.012 0.774	Within cultivar	2.23		
Cultival x woodus	0.012 0.774	Between cultivar	2.81		

Table 5: Harvest index of three cultivars of perennial ryegrass crops sown on two dates and treated with three Moddus[®] rates (a.i. 250 g/l trinexapac-ethyl) in the 2008/09 growing season at Lincoln University, Canterbury.

Discussion

The total seed yield produced was the same for all cultivars but increased when Moddus[®] was applied and when sowing was delayed from 1 April to 14 May (Table 2). Therefore, differences in first grade (saleable) seed yield (Table 3) were associated with the ability of crops to transport assimilate to developing seeds and increase the number of seeds/m² which reached 1.5 mg. 'Grasslands Impact' converted the lowest proportion of total seed to saleable seed at 75% compared with approximately 85% for 'Meridian' and 'Bronsyn'. By definition, the conversion of total seed to first grade seed was through increased individual or TSW. The TSW for 'Grasslands Impact' (2.03 g) was lower than both 'Bronsyn' (2.57 g) and 'Meridian' (2.64 g), which indicates a limitation to seed growth in 'Grasslands Impact'. This suggests the seed yield of 'Grasslands Impact' was sink limited and could be increased if more assimilate was able to be transported to developing seeds.

The application of Moddus[®] increased both total and first grade seed yields up to 1600 ml/ha, with no differences amongst cultivars. None of the treatments imposed affected the number of seed heads/m² or the number of spikelets per head (data not presented). Hence, the total number of spikelets/ m^2 was the same in each crop. Therefore, differences in harvested seed yield arose either from seed weight or the number of seeds/spikelet increasing the number of seeds/m². Moddus[®] had no effect on TSW. The increase in the number of first grade seeds/m² was a result of more seeds/spikelet reaching 1.5 mg. An increase in the number of seeds/spikelet reaching a saleable weight after application of trinexapac-ethyl was also shown for the cultivar 'Cutter' by Chastain et al. (2003). The seed yield increase from the untreated to 1600 ml/ha of Moddus® was linear suggesting higher application rates may increase yields further. Chynoweth et al., (2010), used up to 3.2 l/ha and showed a curvilinear seed yield response up to 3.2 l/ha with reduced lodging.

Delaying sowing from 1 April to 14 May increased first grade seed yield by 545 kg/ha (Table 3). This result contradicts data from Rolston and Archie, (2005) where delayed sowing had a negative effect on harvested seed yield within individual experiments also sown at Lincoln. Rolston and Archie, (2005) used similar sowing dates to this experiment, however, their data showed no effect of sowing date in four cultivars where only the extreme sowing

dates were different from each other. For example, in 2004, sowing on 22 February (1920 kg/ha) showed a higher (P<0.05) seed vield than all other sowing dates, whereas crops sown between 13 March and 30 May were similar (1618 kg/ha). In the current experiment, delaying sowing date to 14 May reduced straw production by 640 kg/ha and concurrently first grade seed weight was increased by 545 kg/ha. Final crop HI was approximately 4% higher for the 14 May sowing date, suggesting a greater proportion of assimilate was allocated towards the seed sink from the delayed sowing date. It is likely that where the same number of potential seed sites were set, spring crop management and environmental conditions were as important as sowing date.

Harvest index (mean 15.0%) was increased by increased first grade seed yield over a constant DM at harvest. Delayed sowing reduced total DM production but increased seed yield, hence these crops had the highest HI (Table 5). In most cases, the application of Moddus® increased HI (12.0-17.6%). However, for 'Grasslands Impact' the non-seed DM/ha at harvest increased when 800 and 1600 ml/ha of Moddus[®] were applied which maintained a constant HI even though seed yield increased. Why DM increased when Moddus[®] was applied for 'Grasslands Impact' only is unclear, but may include greater utilisation of incoming radiation due to reduced lodging (Griffith, 2000). Borm and van den Berg (2008), showed that dry matter production was increased by the application of Moddus[®] when applied later in the season at approximately Zadoks growth stage 33-37 (three nodes visible and tip of final leaf emerging). However, when applied at a single application point no difference in dry matter production was measured.

The overall range of HI (13.5-20%) shown in this experiment was low compared with annual cereal crops (Hay, 1995). Based on the genetic improvements of cereals (Austin et al., 1980), the HI of perennial ryegrass seed crops could be improved through plant breeding and crop management. However, the number of higher order tillers which are produced may limit potential increases in HI. Generally, perennial ryegrass cultivars are selected for vegetative tillering to support pasture dry matter production. As such they produce many more tillers than annual cereal plants. Colvill and Marshall (1984), showed that older, first order tillers produced higher seed yields compared with younger tillers but did not present the weight of stems that produced the seed. Therefore the potential to improve HI may be reduced by the number of second, third and four order tillers which produce seed heads.

The main effects of cultivar, sowing date and Moddus® were all implicated in the absolute lodging percentage at harvest. The application of Moddus[®] reduced stem length by 0.15 m and overall lodging from 95% (horizontal at 0 ml/ha Moddus[®]) to 35% at 1600 ml/ha. These are similar to results presented by Silberstein et al., (2003), Borm and van den Berg (2008) and Rolston et al., (2010). Delaying the sowing date from 1 April to 14 May reduced lodging in 'Bronsyn' and 'Grasslands Impact', but not in 'Meridian' (Table 4) the reasons for cultivar differences are not immediately clear. However, this emphasises the importance of stem shorteners for early sown crops. In cereals, delayed sowing generally reduces the risk of lodging through a reduction in dry matter production (Hay and Walker, 1989). In this experiment dry matter production was 600 kg/ha lower which is insufficient to cause

the difference in lodging when 11,000 kg DM/ha is present (5%). Lodging has been reported to reduce seed yield through reduced assimilate supply to developing seeds (Clemence and Hebblethwaite, 1984), reduced pollination (Wright and Hebblethwaite, 1979) and low seed set (Burbidge et al., 1978), presumably when lodging occurs prior to anthesis. When lodging occurs the canopy has reduced ability to capture and utilise incoming solar which radiation. reduces total photosynthesis capacity. Concurrently maintenance respiration occurs at the same rate in proportion to biomass regardless of canopy integrity. This would lead to less current or stored assimilate available for seed filling (lower yield).

Conclusion

Moddus[®] increased first grade seed yield in three perennial ryegrass cultivars through an increase in the number of seeds/ m^2 which achieved an individual seed weight heavier than 1.5 mg. Moddus® reduced absolute lodging at harvest, shortened stem length and increased crop HI. Growers should consider applying at least 1.6 l/ha to maximise irrigated ryegrass seed Delayed sowing reduced production. lodging in two cultivars and increased first grade seed yield in contrast to previous studies.

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