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Milk production from perennial ryegrass pastures containing different levels of endophyte

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ABSTRACT

These experiments aimed to compare milk production in spring and autumn from perennial ryegrass pastures containing either low (L) (<10%) or high (H) levels of endophyte infection at two pasture allowances. In October 1997 milk yield was higher (P<0.05) from L compared with H endophyte (15.1 v 14.4 kg/cow/day), similarly for fat yield (0.95 v 0.9 kg/cow/day), and milksolids (MS) (1.53 v 1.46 kg/cow/day), but not protein yield. In October 1998 there was no effect of endophyte level on milk, fat, protein and MS yield. In March 1998 milk yield was higher (P<0.001) from H compared to L endophyte (8.5 v 7.4 kg/cow/day), similarly for fat yield (0.53 v 0.48 kg/cow/day), protein yield (0.35 v 0.34 kg/cow/day) and MS yield (0.89 v 0.81 kg/cow/day). No carryover effects were recorded on milk yield or composition in any period.

Keywords: milk yield; perennial ryegrass; endophyte.

INTRODUCTION

New Zealand dairy cows graze pastures containing a high proportion of perennial ryegrass (*Lolium perenne* L.) infected with the fungal endophyte *Neotyphodium lolii*. Chemical compounds, such as ergopeptines and lolitrems, produced by this association have been implicated in heat stress (Easton *et al.,* 1996) and ryegrass staggers (Gallagher *et al.,* 1981). Experiments to test the effect of endophyte-infected ryegrass on milk production from dairy cows have given equivocal results. Valentine *et al.,* (1993) reported that cows grazing pure swards of irrigated endophyte-infected ryegrass in South Australia gave 4-14% less milk than those grazing endophyte-free ryegrass. Butendieck *et al.,* (1994) recorded substantial decreases in milk yield when cut ryegrass (cv Embassy) 100% infected with endophyte was fed after the cultivar Santa Elvira (46% endophyte infection). In this case some cows became recumbent from ryegrass staggers. Where cows were allowed to graze the two cultivars a much smaller decline in milk yield occurred. In New Zealand, Thom *et al.,* (1994) could show no direct relationship between pasture endophyte status and milk production in cows grazing either ryegrass or ryegrass-white clover pastures. Using the same pastures for conserving silage, Clark *et al.,* (1996) showed that endophyte in both pasture and pasture silage had only a transitory effect on milk and protein yield in autumn.

Given the widespread use of endophyte-infected perennial ryegrass for New Zealand dairying even small negative effects on milksolids yield or milk quality may cause large economic losses. Previous work on this topic has not achieved as wide a variation in endophyte-infection as planned because of contamination of pastures established from endophyte-free seed with infected seed transferred in dung and residual surface seed (van Vught & Thom, 1997). This paper reports on three experiments where large contrasts were obtained between endophyte infection levels.

MATERIALS AND METHODS

Treatments and trial design

The three experiments were located at the Dairying Research Corporation, No. 5 Dairy, Hamilton, New Zealand. The planned design was a 2x2 factorial with pasture containing low (L) and high (H) levels of endophyte-infected perennial ryegrass fed at two pasture allowances, nominally 25 and 45 kg DM/cow per day. Experiments 1, 2 and 3 were in spring 1997, autumn 1998 and spring 1998, respectively. Experiments 1 and 3 were as planned, but a severe drought in autumn 1998 meant that only enough pasture was available for one pasture allowance (30 kg DM/cow/day) to be offered. All experimental cows were grazed on spare L endophyte pastures for a week before and after the 2 week experimental periods, the latter was used to determine carryover effects.

Cows

The sixty mixed age cows were grazed in common before the experiments and milk production, liveweight and condition score during this time were used as covariates. Cows were blocked on the basis of breed (experiment 3), age, liveweight, condition score, calving date and milksolids yield and randomly allocated to give fifteen cows per treatment. In experiments 1 and 2 all cows were Jerseys, while in experiment 3 each treatment herd contained 8 Friesians and 7 Jerseys. All cows were drenched daily for bloat with (50 ml 1:6 Bloatenz 2-in-1, Ecolab Ltd) in experiment 1 and 3, and with Zn for facial eczema control in experiment 2. All cows in experiments 1 and 3 were treated with CIDRs to prevent oestrus in cows during the experiments.

MEASUREMENTS

Pasture

Herbage allowances were calculated from calibrated visual assessments of herbage mass made in each paddock before grazing. Calibration samples were cut to ground level with an electric hand piece. Ryegrass tillers

(25 per paddock) were collected for endophyte screening before each experiment to check that maximum possible differences in endophyte levels between treatments occurred in the measurement periods.

Cows

Milk volume and composition (fat, protein and lactose) and somatic cell counts (SCC) were measured on one day in the first week of the experiment, on five days in the second week, and on two days in the post-trial week. Milksolids are defined as fat plus protein. Cows were weighed and scored for condition on 2 consecutive days at the beginning and end of the experiment. All cows were assessed for ryegrass staggers on a weekly basis and assigned a rating of 0-5 based on Keogh (1973). Rectal temperatures were measured by hand-held digital thermometers before afternoon milking at weekly intervals throughout the adjustment, experimental and carryover periods.

Statistical analysis

Analysis of variance and covariance models were used to test treatment differences. Data obtained from individual cow measurements were considered as replicates for analysis of treatment differences. Milk production data were initially analysed as individual test days, but were then pooled on a weekly basis if trends were consistent. Somatic cell count data do not follow a normal distribution and were transformed on a $\log_{10}$ scale before analysis.

RESULTS

In October 1997 (experiment 1) milk yield was higher (P<0.05) for L compared with H endophyte (15.1 vs 14.4 kg/cow per day), similarly for fat yield (0.95 vs 0.90 kg/cow per day) and milksolids yield (1.53 vs 1.46 kg/cow per day), but protein yield was unaffected (Table 1). There were endophyte x allowance effects for fat and protein content (Table 1). Fat content increased from 6.19 to 6.44% with increasing allowance on L endophyte, but decreased from 6.4 to 6.18% with increasing allowance for H endophyte (P<0.05). At low allowance endophyte level had no effect on protein content, but at high allowance protein content for the H endophyte cows was higher than for the L endophyte cows (4.12 vs 3.99%, P<0.001). Cows on L endophyte gained more liveweight (P<0.05) and condition (P<0.05) than those on H endophyte. There was an endophyte x allowance interaction for condition score (P<0.05), with increasing pasture allowance having a larger effect on L compared with H endophyte. As expected, pasture allowance had a highly significant (P<0.001) effect on milk, fat, protein and MS yield as well as liveweight and condition score. No carryover effects were recorded in the week following the experiment. Mean rectal temperatures were 38.5°C and there was no effect of endophyte level. No ryegrass staggers occurred at any stage of the trial.

In March 1998 (experiment 2) milk yield was higher (P<0.001) from H compared with L endophyte (8.5 vs 7.4) kg/cow per day, similarly for fat yield (0.53 vs 0.48 kg/cow per day), protein (0.35 vs 0.34 kg/cow per day) and milksolids (0.89 vs 0.81 kg/cow per day) (Table 2). There was no effect of endophyte level on milk composition. The only carryover effect recorded was a higher (P<0.05) protein yield for the H endophyte cow in the week following the experiment. Cows maintained liveweight and condition score over the trial period of two weeks and there were no effects of endophyte level. Mean rectal temperatures were 38.6°C and there was no effect of endophyte level. Several cows on the H endophyte showed signs of intermittent ryegrass staggers but were never scored above 1 in severity, no staggers was recorded on the L endophyte pastures.

In October 1998 (experiment 3) there was no effect of endophyte level on milk, fat, protein and MS yield (Table 3). Protein content was increased (P<0.05) on the H endophyte pastures (3.67 vs 3.58%). In contrast, lactose content was higher (P<0.05) on the L endophyte pastures (5.09 vs 5.04%). Somatic cell counts on all treatments were very low with an overall mean of 47,500 cells/ml. Only on one of the five measurement days was a significant treatment difference recorded, in this case the H endophyte cows had lower (P<0.05) SCC than the L endophyte (39,600 vs 50,100 cells/ml). Cows on all treatments gained liveweight and condition score and there were no treatment effects on these variables. Mean rectal temperatures were 38.4°C with no effect of endophyte level. No ryegrass staggers occurred at any stage of the trial. The low levels of ergovaline and lolitrem B recorded in the experimental periods (Table 4) may explain the lack of treatment effect on rectal temperatures and ryegrass staggers. As expected, pasture allowance had a highly significant effect (P<0.001) on milk, fat, protein and MS yield. No carryover effects were recorded in the week following the experiment.

TABLE 1: Adjusted mean milk, fat, protein and milksolids yield and milk composition from cows grazing high (H) or Low (L) endophyte ryegrass pastures at pasture allowances of 25 or 45 kg DM/cow per day (experiment 1 - spring 1997).

Treatment		Milk	Fat	Protein	Milksolids	Fat	Protein	Lactose
Endophyte	Allowance	(kg/cow per day)				(%)		
H	25	12.4	0.79	0.45	1.24	6.40	3.63	5.20
H	45	16.4	1.01	0.68	1.69	6.18	4.12	5.20
L	25	13.5	0.83	0.50	1.32	6.19	3.69	5.22
L	45	16.6	1.07	0.66	1.74	6.44	3.99	5.21
SED (main effects)		0.28	0.025	0.011	0.034	0.107	0.024	0.018
SED (Interactions)		0.41	0.035	0.016	0.048	0.155	0.034	0.025

TABLE 2: Adjusted mean milk, fat, protein and milksolids yield and milk composition from cows grazing high (H) or Low (L) endophyte ryegrass pastures (experiment 2 - autumn 1998).

Treatment	Milk	Fat	Protein	Milksolids	Fat	Protein	Lactose
Endophyte	(kg/cow per day)				(%)		
H	8.5	0.53	0.35	0.89	6.36	4.25	4.80
L	7.4	0.48	0.34	0.81	6.16	4.21	4.78
SED	0.17	0.012	0.006	0.017	0.11	0.038	0.016

TABLE 3: Adjusted mean milk, fat, protein and milksolids yield and milk composition from cows grazing high (H) or Low (L) endophyte ryegrass pastures at pasture allowances of 25 or 45 kg DM/cow per day (experiment 3 - spring 1998).

Treatment		Milk	Fat	Protein	Milksolids	Fat	Protein	Lactose
Endophyte	Allowance	(kg/cow per day)				(%)		
H	25	17.8	0.84	0.64	1.47	4.79	3.64	5.05
H	45	19.9	0.94	0.73	1.67	4.81	3.70	5.03
L	25	18.1	0.85	0.63	1.48	4.78	3.54	5.10
L	45	19.7	0.93	0.70	1.64	4.88	3.62	5.07
SED(Main effects)		0.35	0.02	0.013	0.035	0.09	0.039	0.016

DISCUSSION

The higher milk yield from L compared with H endophyte pastures in spring 1997 contrasts with Thom *et al*., (1994) who reported no effect of endophyte on spring milk production. However, no endophyte effect was present in spring 1998. In the 1997 experiment lolitrem B levels on H endophyte were low, and non-detectable on L endophyte; paxilline and lysergol (Table 4) were not detected in either treatment. Therefore, the difference in milk yield between the two experiments is unexpected. It is likely that intakes were higher on the L endophyte pastures because the higher milk yield was associated with both higher liveweight gain and condition score. In neither year was protein yield affected by endophyte level, because in 1997 the lower milk yield on H endophyte pastures was compensated for by higher milk protein content. We are able to offer no explanation for either the effect on milk composition in both years, or the endophyte x allowance effects in 1997. Endophyte effects on milk composition were not detected by Strahan *et al*., (1987), Valentine *et al*., (1993) or Bernard *et al*., (1993).

TABLE 4: Lolitrem B, Paxilline, Lysergol and Ergovaline levels for perennial ryegrass at the two experimental times. (Data for Experiment 3 are unavailable).

Treatment Endophyte	Allowance	Lolitrem B	Paxilline (μg/g)	Lysergol	Ergovaline
Experiment 1					
H	25	0.20	nd	nd	0.3
H	45	0.18	nd	nd	0.3
L	25	nd	nd	nd	0
L	45	nd	nd	nd	0
Experiment 2					
H		1.72	3.02	0.014	0.1
L		0.62	1.02	0.007	0.1

nd = not detectable

Anecdotal evidence from farmers suggests that high endophyte pastures may be associated with high somatic cell count. The results from spring 1998 show that at least in circumstances where endophyte levels are not affecting milk yields there is no effect on SCC. This agrees with previous work at this site (M. J. Auldist, pers. comm.). No published information on the effect of endophyte on SCC could be found.

The autumn 1998 result where H endophyte gave higher milk yield than L endophyte was unexpected. Previous work (Valentine *et al*., 1993, Clark *et al*., 1996) on autumn pastures suggests that this is the most likely time for deleterious effects to occur. Air temperatures are high and USA studies on tall fescue endophyte showed that effects are greatest above 25-30°C (Hemken *et al.,* 1981). Ergovaline levels were shown to peak in New Zealand pastures in February-March (Easton *et al*., 1996). Lolitrem B levels also reach a peak at this time (Thom *et al*., 1994). In the present experiment lolitrem B levels were 1.72 mg/g DM on the H endophyte. Only intermittent ryegrass staggers of low severity was recorded which is consistent with the observation that > 2μg/g DM of lolitrem B is required for ryegrass staggers to occur (Prestidge & Gallagher, 1989). Reduced levels of green ryegrass leaf in autumn (Thom *et al*., 1994) mean that cows are likely to ingest greater amounts of ryegrass pseudostem material which has a higher concentration of ergovaline and lolitrem B than leaf (Keogh & Tapper, 1993).

There is ample evidence that endophyte-infected tall fescue eaten by dairy cows can cause decreased milk yield compared with endophyte-free tall fescue (Strahan *et al*., 1987, Hemken *et al*., 1979). However, there are also instances where no decrease in milk yield has occurred (Jackson *et al*., 1997). The largest increases in milk yield (20-35%) from low endophyte tall fescue or perennial ryegrass compared with high endophyte pastures have been reported when cut pastures have been fed to cows in stalls (Strahan *et al*., 1987; Butendieck *et al.,* 1994). This method of harvesting and feeding greatly reduces the opportunity for diet selection and may increase the amount of high endophyte material fed compared to a grazed comparison. Therefore, extrapolation from stall feeding experiments to grazing should be done with caution.

Although high ergopeptine levels have been associated with decreased milk yield there is no unequivocal evidence that ergovaline or other related compounds are the causative agents.

In a comprehensive review of endophyte toxicosis in ruminants, Oliver (1997) states "dose response studies of specific alkaloids must be made to confirm individual alkaloid effect". This has not been done for ergovaline or any other endophyte-derived compound in dairy cows. An excellent example of the dangers of assuming ergovaline to be the major causative agent of endophyte toxicosis is given by Piper *et al*. (1997). They fed rats endophyte-free seed plus ergovaline compared with endophyte-infected seed containing an equivalent amount of ergovaline. The former treatment did not change pre-treatment levels of feed intake, liveweight gain or serum prolactin, whereas the latter gave significant reductions in all response parameters. The results strongly suggest that endophyte-derived compounds other than ergovaline play an important role in producing endophyte toxicosis symptoms.

Strahan *et al*., (1987) recorded increased rectal temperatures associated with decreased milk yield for cows fed high endophyte tall fescue. In the present experiments there were never any treatment effects on rectal temperature which suggests that ergovaline and/or other compounds were not present in sufficient quantities to disturb homeostatic mechanisms.

Stuedemann *et al.,* (1989) compared beef cattle intake and growth on endophyte-infected and endophyte-free pasture and found carryover effects on intake for at least 10 days after all animals were removed to endophyte-free pastures, with intakes returned to normal 28 days after removal. In the present spring 1997 experiment no carryover effects of endophyte level on milk yield or composition were noted, suggesting that cows quickly recovered from any deleterious effect.

The present experiment and that of Valentine *et al*., (1993) are the two major grazing trials comparing perennial ryegrass pastures with widely different endophyte levels and unconfounded with white clover levels. The spring result from Valentine *et al.,* (1993) showed a significant effect on milk yield of 4.5%, the same as for our spring 1997 experiment, but in the former neither protein nor fat yield were significantly different, whereas in the present experiment milksolids differences reflected milk yield differences. The March results were quite different for the two experiments. Ergovaline levels are not reported by Valentine *et al*., (1993), but our March 1997 results, show ergovaline levels of 0.1μ/g DM for both treatments (Table 4). These are much lower than the mean value for ergovaline of 1.5 μg/g DM (range 0.5 – 2.0 μg/g DM) that Easton *et al*., (1996) found in February – March for 48 samples taken throughout New Zealand. A partial explanation may be the lower levels of ryegrass in the sward because of volunteer summer grasses (Table 5) which are often preferentially grazed because of their high green leaf content. This suggests that it will not be possible to generalise about the effects of endophyte on milk yield from grazed pastures. Much more specific information is likely to be needed from pasture, animal and environmental perspectives.

TABLE 5: Botanical composition of treatment pastures at the three experimental times.

Treatment Endophyte	Allowance	Ryegrass	White Clover	Poa spp.	C4 grasses* (%)	Other grasses	Weeds	Dead
Experiment 1								
H	25	77.3	1.7	16.5	0	1.1	1.0	2.4
H	45	84.9	2.8	7.6	0	0	0.1	4.6
L	25	79.8	1.2	10.3	0	0.1	2.9	5.7
L	45	74.9	4.7	14.7	0	0	0.4	5.3
Experiment 2								
H		37.8	3.7	0	33.5	0	3.0	22.0
L		43.5	1.1	0	9.3	9.5	3.9	32.7
Experiment 3								
H	25	73.4	7.7	9.8	0	0	1.0	8.1
H	45	76.4	4.0	9.9	0	0	3.9	5.8
L	25	75.8	0	10.9	0	0	7.5	5.8
l	45	84.2	0	9.4	0	0	2.5	3.9

* Predominantly summer grass (*Digitaria sanguinalis*) and smooth witchgrass (*Panicum dichotomiflorum*).

PRACTICAL IMPLICATIONS

The significant decline in MS yield of 4.25% on H endophyte pastures in spring 1997, needs to be set against the lack of effect in spring 1998 and the positive effect in autumn 1998, and the lack of effect on SCC, ryegrass staggers and heat stress. High endophyte pastures provide partial protection against insect damage, and it would be unwise for dairy farmers in general to replace H endophyte with L endophyte pastures. Where heat stress and/or ryegrass staggers are major, recurring problems there may be some merit in replacing a proportion of the H endophyte pastures with pasture species that don't contain endophyte. However, recent research by van Vught & Thom (1997) has clearly shown that contamination of new pastures will occur rapidly from surviving ryegrass remnants and from H endophyte seeds surviving in dung in many circumstances.

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Keywords: milk yield; perennial ryegrass; endophyte.

INTRODUCTION

New Zealand dairy cows graze pastures containing a high proportion of perennial ryegrass (*Lolium perenne* L.) infected with the fungal endophyte *Neotyphodium lolii*. Chemical compounds, such as ergopeptines and lolitrems, produced by this association have been implicated in heat stress (Easton *et al.,* 1996) and ryegrass staggers (Gallagher *et al.,* 1981). Experiments to test the effect of endophyte-infected ryegrass on milk production from dairy cows have given equivocal results. Valentine *et al.,* (1993) reported that cows grazing pure swards of irrigated endophyte-infected ryegrass in South Australia gave 4-14% less milk than those grazing endophyte-free ryegrass. Butendieck *et al.,* (1994) recorded substantial decreases in milk yield when cut ryegrass (cv Embassy) 100% infected with endophyte was fed after the cultivar Santa Elvira (46% endophyte infection). In this case some cows became recumbent from ryegrass staggers. Where cows were allowed to graze the two cultivars a much smaller decline in milk yield occurred. In New Zealand, Thom *et al.,* (1994) could show no direct relationship between pasture endophyte status and milk production in cows grazing either ryegrass or ryegrass-white clover pastures. Using the same pastures for conserving silage, Clark *et al.,* (1996) showed that endophyte in both pasture and pasture silage had only a transitory effect on milk and protein yield in autumn.

Given the widespread use of endophyte-infected perennial ryegrass for New Zealand dairying even small negative effects on milksolids yield or milk quality may cause large economic losses. Previous work on this topic has not achieved as wide a variation in endophyte-infection as planned because of contamination of pastures established from endophyte-free seed with infected seed transferred in dung and residual surface seed (van Vught & Thom, 1997). This paper reports on three experiments where large contrasts were obtained between endophyte infection levels.

MATERIALS AND METHODS

Treatments and trial design

The three experiments were located at the Dairying Research Corporation, No. 5 Dairy, Hamilton, New Zealand. The planned design was a 2x2 factorial with pasture containing low (L) and high (H) levels of endophyte-infected perennial ryegrass fed at two pasture allowances, nominally 25 and 45 kg DM/cow per day. Experiments 1, 2 and 3 were in spring 1997, autumn 1998 and spring 1998, respectively. Experiments 1 and 3 were as planned, but a severe drought in autumn 1998 meant that only enough pasture was available for one pasture allowance (30 kg DM/cow/day) to be offered. All experimental cows were grazed on spare L endophyte pastures for a week before and after the 2 week experimental periods, the latter was used to determine carryover effects.

Cows

The sixty mixed age cows were grazed in common before the experiments and milk production, liveweight and condition score during this time were used as covariates. Cows were blocked on the basis of breed (experiment 3), age, liveweight, condition score, calving date and milksolids yield and randomly allocated to give fifteen cows per treatment. In experiments 1 and 2 all cows were Jerseys, while in experiment 3 each treatment herd contained 8 Friesians and 7 Jerseys. All cows were drenched daily for bloat with (50 ml 1:6 Bloatenz 2-in-1, Ecolab Ltd) in experiment 1 and 3, and with Zn for facial eczema control in experiment 2. All cows in experiments 1 and 3 were treated with CIDRs to prevent oestrus in cows during the experiments.

MEASUREMENTS

Pasture

Herbage allowances were calculated from calibrated visual assessments of herbage mass made in each paddock before grazing. Calibration samples were cut to ground level with an electric hand piece. Ryegrass tillers

(25 per paddock) were collected for endophyte screening before each experiment to check that maximum possible differences in endophyte levels between treatments occurred in the measurement periods.

Cows

Milk volume and composition (fat, protein and lactose) and somatic cell counts (SCC) were measured on one day in the first week of the experiment, on five days in the second week, and on two days in the post-trial week. Milksolids are defined as fat plus protein. Cows were weighed and scored for condition on 2 consecutive days at the beginning and end of the experiment. All cows were assessed for ryegrass staggers on a weekly basis and assigned a rating of 0-5 based on Keogh (1973). Rectal temperatures were measured by hand-held digital thermometers before afternoon milking at weekly intervals throughout the adjustment, experimental and carryover periods.

Statistical analysis

Analysis of variance and covariance models were used to test treatment differences. Data obtained from individual cow measurements were considered as replicates for analysis of treatment differences. Milk production data were initially analysed as individual test days, but were then pooled on a weekly basis if trends were consistent. Somatic cell count data do not follow a normal distribution and were transformed on a $\log_{10}$ scale before analysis.

RESULTS

In October 1997 (experiment 1) milk yield was higher (P<0.05) for L compared with H endophyte (15.1 vs 14.4 kg/cow per day), similarly for fat yield (0.95 vs 0.90 kg/cow per day) and milksolids yield (1.53 vs 1.46 kg/cow per day), but protein yield was unaffected (Table 1). There were endophyte x allowance effects for fat and protein content (Table 1). Fat content increased from 6.19 to 6.44% with increasing allowance on L endophyte, but decreased from 6.4 to 6.18% with increasing allowance for H endophyte (P<0.05). At low allowance endophyte level had no effect on protein content, but at high allowance protein content for the H endophyte cows was higher than for the L endophyte cows (4.12 vs 3.99%, P<0.001). Cows on L endophyte gained more liveweight (P<0.05) and condition (P<0.05) than those on H endophyte. There was an endophyte x allowance interaction for condition score (P<0.05), with increasing pasture allowance having a larger effect on L compared with H endophyte. As expected, pasture allowance had a highly significant (P<0.001) effect on milk, fat, protein and MS yield as well as liveweight and condition score. No carryover effects were recorded in the week following the experiment. Mean rectal temperatures were 38.5°C and there was no effect of endophyte level. No ryegrass staggers occurred at any stage of the trial.

In March 1998 (experiment 2) milk yield was higher (P<0.001) from H compared with L endophyte (8.5 vs 7.4) kg/cow per day, similarly for fat yield (0.53 vs 0.48 kg/cow per day), protein (0.35 vs 0.34 kg/cow per day) and milksolids (0.89 vs 0.81 kg/cow per day) (Table 2). There was no effect of endophyte level on milk composition. The only carryover effect recorded was a higher (P<0.05) protein yield for the H endophyte cow in the week following the experiment. Cows maintained liveweight and condition score over the trial period of two weeks and there were no effects of endophyte level. Mean rectal temperatures were 38.6°C and there was no effect of endophyte level. Several cows on the H endophyte showed signs of intermittent ryegrass staggers but were never scored above 1 in severity, no staggers was recorded on the L endophyte pastures.

In October 1998 (experiment 3) there was no effect of endophyte level on milk, fat, protein and MS yield (Table 3). Protein content was increased (P<0.05) on the H endophyte pastures (3.67 vs 3.58%). In contrast, lactose content was higher (P<0.05) on the L endophyte pastures (5.09 vs 5.04%). Somatic cell counts on all treatments were very low with an overall mean of 47,500 cells/ml. Only on one of the five measurement days was a significant treatment difference recorded, in this case the H endophyte cows had lower (P<0.05) SCC than the L endophyte (39,600 vs 50,100 cells/ml). Cows on all treatments gained liveweight and condition score and there were no treatment effects on these variables. Mean rectal temperatures were 38.4°C with no effect of endophyte level. No ryegrass staggers occurred at any stage of the trial. The low levels of ergovaline and lolitrem B recorded in the experimental periods (Table 4) may explain the lack of treatment effect on rectal temperatures and ryegrass staggers. As expected, pasture allowance had a highly significant effect (P<0.001) on milk, fat, protein and MS yield. No carryover effects were recorded in the week following the experiment.

TABLE 1: Adjusted mean milk, fat, protein and milksolids yield and milk composition from cows grazing high (H) or Low (L) endophyte ryegrass pastures at pasture allowances of 25 or 45 kg DM/cow per day (experiment 1 - spring 1997).

Treatment		Milk	Fat	Protein	Milksolids	Fat	Protein	Lactose
Endophyte	Allowance	(kg/cow per day)				(%)		
H	25	12.4	0.79	0.45	1.24	6.40	3.63	5.20
H	45	16.4	1.01	0.68	1.69	6.18	4.12	5.20
L	25	13.5	0.83	0.50	1.32	6.19	3.69	5.22
L	45	16.6	1.07	0.66	1.74	6.44	3.99	5.21
SED (main effects)		0.28	0.025	0.011	0.034	0.107	0.024	0.018
SED (Interactions)		0.41	0.035	0.016	0.048	0.155	0.034	0.025

TABLE 2: Adjusted mean milk, fat, protein and milksolids yield and milk composition from cows grazing high (H) or Low (L) endophyte ryegrass pastures (experiment 2 - autumn 1998).

Treatment	Milk	Fat	Protein	Milksolids	Fat	Protein	Lactose
Endophyte	(kg/cow per day)				(%)		
H	8.5	0.53	0.35	0.89	6.36	4.25	4.80
L	7.4	0.48	0.34	0.81	6.16	4.21	4.78
SED	0.17	0.012	0.006	0.017	0.11	0.038	0.016

TABLE 3: Adjusted mean milk, fat, protein and milksolids yield and milk composition from cows grazing high (H) or Low (L) endophyte ryegrass pastures at pasture allowances of 25 or 45 kg DM/cow per day (experiment 3 - spring 1998).

Treatment		Milk	Fat	Protein	Milksolids	Fat	Protein	Lactose
Endophyte	Allowance	(kg/cow per day)				(%)		
H	25	17.8	0.84	0.64	1.47	4.79	3.64	5.05
H	45	19.9	0.94	0.73	1.67	4.81	3.70	5.03
L	25	18.1	0.85	0.63	1.48	4.78	3.54	5.10
L	45	19.7	0.93	0.70	1.64	4.88	3.62	5.07
SED(Main effects)		0.35	0.02	0.013	0.035	0.09	0.039	0.016

DISCUSSION

The higher milk yield from L compared with H endophyte pastures in spring 1997 contrasts with Thom *et al*., (1994) who reported no effect of endophyte on spring milk production. However, no endophyte effect was present in spring 1998. In the 1997 experiment lolitrem B levels on H endophyte were low, and non-detectable on L endophyte; paxilline and lysergol (Table 4) were not detected in either treatment. Therefore, the difference in milk yield between the two experiments is unexpected. It is likely that intakes were higher on the L endophyte pastures because the higher milk yield was associated with both higher liveweight gain and condition score. In neither year was protein yield affected by endophyte level, because in 1997 the lower milk yield on H endophyte pastures was compensated for by higher milk protein content. We are able to offer no explanation for either the effect on milk composition in both years, or the endophyte x allowance effects in 1997. Endophyte effects on milk composition were not detected by Strahan *et al*., (1987), Valentine *et al*., (1993) or Bernard *et al*., (1993).

TABLE 4: Lolitrem B, Paxilline, Lysergol and Ergovaline levels for perennial ryegrass at the two experimental times. (Data for Experiment 3 are unavailable).

Treatment Endophyte	Allowance	Lolitrem B	Paxilline (µg/g)	Lysergol	Ergovaline
Experiment 1					
H	25	0.20	nd	nd	0.3
H	45	0.18	nd	nd	0.3
L	25	nd	nd	nd	0
L	45	nd	nd	nd	0
Experiment 2					
H		1.72	3.02	0.014	0.1
L		0.62	1.02	0.007	0.1

nd = not detectable

Anecdotal evidence from farmers suggests that high endophyte pastures may be associated with high somatic cell count. The results from spring 1998 show that at least in circumstances where endophyte levels are not affecting milk yields there is no effect on SCC. This agrees with previous work at this site (M. J. Auldist, pers. comm.). No published information on the effect of endophyte on SCC could be found.

The autumn 1998 result where H endophyte gave higher milk yield than L endophyte was unexpected. Previous work (Valentine *et al*., 1993, Clark *et al*., 1996) on autumn pastures suggests that this is the most likely time for deleterious effects to occur. Air temperatures are high and USA studies on tall fescue endophyte showed that effects are greatest above 25-30°C (Hemken *et al.,* 1981). Ergovaline levels were shown to peak in New Zealand pastures in February-March (Easton *et al*., 1996). Lolitrem B levels also reach a peak at this time (Thom *et al*., 1994). In the present experiment lolitrem B levels were 1.72 mg/g DM on the H endophyte. Only intermittent ryegrass staggers of low severity was recorded which is consistent with the observation that > 2µg/g DM of lolitrem B is required for ryegrass staggers to occur (Prestidge & Gallagher, 1989). Reduced levels of green ryegrass leaf in autumn (Thom *et al*., 1994) mean that cows are likely to ingest greater amounts of ryegrass pseudostem material which has a higher concentration of ergovaline and lolitrem B than leaf (Keogh & Tapper, 1993).

There is ample evidence that endophyte-infected tall fescue eaten by dairy cows can cause decreased milk yield compared with endophyte-free tall fescue (Strahan *et al*., 1987, Hemken *et al*., 1979). However, there are also instances where no decrease in milk yield has occurred (Jackson *et al*., 1997). The largest increases in milk yield (20-35%) from low endophyte tall fescue or perennial ryegrass compared with high endophyte pastures have been reported when cut pastures have been fed to cows in stalls (Strahan *et al*., 1987; Butendieck *et al.,* 1994). This method of harvesting and feeding greatly reduces the opportunity for diet selection and may increase the amount of high endophyte material fed compared to a grazed comparison. Therefore, extrapolation from stall feeding experiments to grazing should be done with caution.

Although high ergopeptine levels have been associated with decreased milk yield there is no unequivocal evidence that ergovaline or other related compounds are the causative agents.

In a comprehensive review of endophyte toxicosis in ruminants, Oliver (1997) states "dose response studies of specific alkaloids must be made to confirm individual alkaloid effect". This has not been done for ergovaline or any other endophyte-derived compound in dairy cows. An excellent example of the dangers of assuming ergovaline to be the major causative agent of endophyte toxicosis is given by Piper *et al*. (1997). They fed rats endophyte-free seed plus ergovaline compared with endophyte-infected seed containing an equivalent amount of ergovaline. The former treatment did not change pre-treatment levels of feed intake, liveweight gain or serum prolactin, whereas the latter gave significant reductions in all response parameters. The results strongly suggest that endophyte-derived compounds other than ergovaline play an important role in producing endophyte toxicosis symptoms.

Strahan *et al*., (1987) recorded increased rectal temperatures associated with decreased milk yield for cows fed high endophyte tall fescue. In the present experiments there were never any treatment effects on rectal temperature which suggests that ergovaline and/or other compounds were not present in sufficient quantities to disturb homeostatic mechanisms.

Stuedemann *et al.,* (1989) compared beef cattle intake and growth on endophyte-infected and endophyte-free pasture and found carryover effects on intake for at least 10 days after all animals were removed to endophyte-free pastures, with intakes returned to normal 28 days after removal. In the present spring 1997 experiment no carryover effects of endophyte level on milk yield or composition were noted, suggesting that cows quickly recovered from any deleterious effect.

The present experiment and that of Valentine *et al*., (1993) are the two major grazing trials comparing perennial ryegrass pastures with widely different endophyte levels and unconfounded with white clover levels. The spring result from Valentine *et al.,* (1993) showed a significant effect on milk yield of 4.5%, the same as for our spring 1997 experiment, but in the former neither protein nor fat yield were significantly different, whereas in the present experiment milksolids differences reflected milk yield differences. The March results were quite different for the two experiments. Ergovaline levels are not reported by Valentine *et al*., (1993), but our March 1997 results, show ergovaline levels of 0.1μ/g DM for both treatments (Table 4). These are much lower than the mean value for ergovaline of 1.5 μg/g DM (range 0.5 – 2.0 μg/g DM) that Easton *et al*., (1996) found in February – March for 48 samples taken throughout New Zealand. A partial explanation may be the lower levels of ryegrass in the sward because of volunteer summer grasses (Table 5) which are often preferentially grazed because of their high green leaf content. This suggests that it will not be possible to generalise about the effects of endophyte on milk yield from grazed pastures. Much more specific information is likely to be needed from pasture, animal and environmental perspectives.

TABLE 5: Botanical composition of treatment pastures at the three experimental times.

Treatment Endophyte	Allowance	Ryegrass	White Clover	Poa spp.	C4 grasses* (%)	Other grasses	Weeds	Dead
Experiment 1								
H	25	77.3	1.7	16.5	0	1.1	1.0	2.4
H	45	84.9	2.8	7.6	0	0	0.1	4.6
L	25	79.8	1.2	10.3	0	0.1	2.9	5.7
L	45	74.9	4.7	14.7	0	0	0.4	5.3
Experiment 2								
H		37.8	3.7	0	33.5	0	3.0	22.0
L		43.5	1.1	0	9.3	9.5	3.9	32.7
Experiment 3								
H	25	73.4	7.7	9.8	0	0	1.0	8.1
H	45	76.4	4.0	9.9	0	0	3.9	5.8
L	25	75.8	0	10.9	0	0	7.5	5.8
l	45	84.2	0	9.4	0	0	2.5	3.9

* Predominantly summer grass (*Digitaria sanguinalis*) and smooth witchgrass (*Panicum dichotomiflorum*).

PRACTICAL IMPLICATIONS

The significant decline in MS yield of 4.25% on H endophyte pastures in spring 1997, needs to be set against the lack of effect in spring 1998 and the positive effect in autumn 1998, and the lack of effect on SCC, ryegrass staggers and heat stress. High endophyte pastures provide partial protection against insect damage, and it would be unwise for dairy farmers in general to replace H endophyte with L endophyte pastures. Where heat stress and/or ryegrass staggers are major, recurring problems there may be some merit in replacing a proportion of the H endophyte pastures with pasture species that don't contain endophyte. However, recent research by van Vught & Thom (1997) has clearly shown that contamination of new pastures will occur rapidly from surviving ryegrass remnants and from H endophyte seeds surviving in dung in many circumstances.

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