Effect of perennial ryegrass endophyte and a feed additive on some physiological parameters and intake of young ewes in winter

M L.E. HENRY[1], M. HAZELTON[2], K.F.M. REED[3], L.J. CUMMINS[4] and B.J. LEURY[1]
[1] *School of Agriculture and Food Systems, University of Melbourne, Parkville, Vic.*
[2] *Feedworks Pty Ltd, Romsey, Vic., Australia*
[3] *Reed Pasture Science, Hamilton, Vic., Australia*
[4] *Ivanhoe*, Cavendish, *Vic., Australia*
brianjl@unimelb.edu.au

Abstract

Young merino ewes were offered a pelleted ration containing approx 15% perennial ryegrass seed, either infected or not infected with wild endophyte. Each group was split so that 10 ewes received the toxin deactivation product, Elitox®. Groups were fed indoors on treatment for 4 weeks in winter (average daily temperature, 11.5ºC).

Analysis for endophyte toxins indicated intakes of 63 µg/kg LW/day ergovaline and 23 µg/kg LW/day lolitrem B. Increases in respiration rate and rectal temperature were obvious after 1 week. Respiration rate more than doubled for ewes ingesting toxin-containing wild endophyte. Differences were greater with observations taken at 9 pm, compared with those taken at 9 am. The intake and liveweight gain of ewes ingesting endophyte increased relative to those on the endophyte-free diet. Administration of Elitox®, resulted in a significant interaction: respiration rate and intake increased in the absence of endophyte, but decreased in its presence.

Keywords: Merino sheep, perennial ryegrass, lolitrem B, ergovaline, Elitox®, intake, respiration rate, faecal moisture

Introduction

Perennial ryegrass endophyte toxicosis (PRGT) is characterised by staggers and heat stress, effects on blood hormones, liveweight and reproduction. Toxins include lolitrem B and ergovaline. Lolitrem B causes endocrine dysfunction, and respiratory, cardiovascular, thermoregulatory and digestive problems. Ergovaline decreases milk production and disrupts thermoregulation and endocrine function (Oliver 2005).

Wild endophyte in ryegrass typically increases the rectal temperature of sheep in summer/autumn and raises respiration rate (Fletcher *et al*. 1999). Negative effects on the growth of

Table 1 Intake of dry matter (g/hd/day) and toxin (µg/kg LW/day), LW (kg), ADG (g) and faecal moisture (%) for each group.

						---------- P ----------		
	C	CElitox	T	TElitox	SED	T	Elitox	T x Elitox
Intake of dry matter								
Baseline	619[a]	628[a]	630[a]	559[b]	59.2	0.463	0.434	0.318
Week 1,2	776[a]	858[ab]	940[b]	774[a]	101.3	0.005	0.323	0.010
Week 3,4	632[a]	1037[b]	927[b]	907[b]	120.8	0.064	0.057	0.135
Intake of ergovaline								
Week 1,2	-	-	65[a]	42[b]	4.6	-	0.000	-
Week 3,4	-	-	58[a]	50[a]	5.9	-	0.157	-
Intake of lolitrem B								
Week 1,2	-	-	25[a]	15[b]	1.7	-	0.000	-
Week 3,4	-	-	22[a]	18[b]	2.1	-	0.042	-
Liveweight								
Initial	31.0	30.6	31.4	30.4	1.10	0.928	0.349	0.701
Final [c]	33.0	34.9	34.8	33.1	0.94	0.974	0.857	0.006
Average daily gain								
Weeks1-2	107[ab]	127[a]	157[a]	50[b]	31.8	0.069	0.004	0.000
Weeks 2-4	44[a]	161[b]	131[ab]	160[b]	49.5	0.199	0.035	0.198
Weeks 1-4	66[a]	146[b]	142[b]	81[ab]	33.3	0.807	0.682	0.003
Faecal moisture								
Day 28	74.9	77.2	77.7	78.5	2.7	0.312	0.440	0.761

[a, b,] Means with different superscripts are different (P<0.05)
[c] initial LW as a covariate

sheep grazing infected perennial ryegrass compared to those grazing non-infected ryegrass were found by Watson *et al.* (1999) and Cosgrove & Hume (2005). Decreased consumption was attributed to lower acceptance. Foot *et al*. (1994) found increased faecal moisture in sheep grazing wild endophyte-infected perennial ryegrass; fly strike may also be greater (Young *et al*. 2004).

Elitox® was designed by Impextraco (Belgium) to bind and inactivate mycotoxins. Ingredients include a natural biopolymer (chitosan), enzymes, natural extracts, vitamin-C and carriers including hydrated sodium calcium aluminium silicates. Our study aimed to (1) determine the effect on animal production and physiology from feeding a known concentration of ergovaline and lolitrem B in the diet of merino ewe weaners and (2) determine whether Elitox® is able to ameliorate the effect of toxin ingestion.

Materials and Method

Merino ewe weaners (9 months old) were kept in an outdoor pen for 14 days to acclimatise to pelleted rations. Forty were selected on liveweight (LW) and allocated to one of four indoor pen-feeding systems. Each pen (4 x 6 m) contained an automatic feeder with tag reader enabling records of individuals' meals. The control diet [C] contained seed of perennial ryegrass that was endophyte free. The toxin diet [T] which contained seed of wild endophyte-infected perennial ryegrass, contained ergovaline (1.9 mg/kg) and lolitrem B (0.7 mg/kg). Both C and T diets had Elitox® added to make up the other two treatments, CElitox and TElitox. Pellets (79% DOMD, 19% crude protein, 13.5 MJ/kg DM) contained pea meal, wheat, perennial ryegrass seed, canola, lucerne chaff, molasses, acid buff, lime, ammonium chloride, salt and sheep premix. On moving indoors, sheep pellets were fed for 9 days, then the control pellet ration [C] was fed for the last 10 days of acclimatisation. Straw was fed daily (1 kg/pen). The four groups were then fed the different diets over 28 days. At completion of the experiment a composite sample of each feed collected during the experiment was analysed for nutritive value, ergovaline and lolitrem B.

Sheep were weighed and their respiration rate and rectal temperature was recorded three times per week at 9.00am and 9.00pm. On the second last day of the experiment animals were put under an acute exercise regime for 10 minutes, then rested for 30 minutes before taking observations. Faecal samples were taken at week four to determine moisture content.

Generalised linear models were generated using covariate analysis where appropriate. Regression analysis was used to determine if there was any relationship between toxin ingestion and the physiological parameters.

Results

Daily air temperature inside the animal house ranged from 1-11ºC on a minimum basis and from 12-23ºC on a maximum basis. Average daily temperature was 11.5ºC. T ewes ingested approximately 63 µg/kg LW/day ergovaline and 23 µg/kg LW/day lolitrem B. Their dry matter intake (DMI) and average daily gain (ADG) was greater than for ewes fed C (Table 1). Averaged over Weeks 1-4, toxin ingestion (T) increased respiration by approximately 50 breaths/minute and rectal temperature by approximately 0.4ºC compared with group C (Table 2). During the first fortnight significant interactions occurred for DMI and ADG: the addition of Elitox® increased DMI and ADG for C sheep and decreased DMI and ADG for T sheep. Sheep

Table 2 Respiration rate (breaths/min) and rectal temperature (ºC) averaged over 24 hour periods during the feeding experiment and pre and post-exercise.

						---------- P ----------		
	C	CElitox	T	TElitox	SED	T	Elitox	T x Elitox
Respiration rate								
Week 0	47	54	53	46	3.7	0.585	0.972	0.008
Week 1	66	69	89	70	12.2	0.143	0.343	0.169
Week 2	38[a]	70[b]	87[c]	40[a]	6.4	0.028	0.103	0.000
Week 3	30[a]	77[b]	93[c]	77[b]	11.2	0.000	0.045	0.000
Week 4	42[a]	75[b]	97[b]	77[b]	10.9	0.000	0.414	0.001
Rectal temperature								
Baseline	39.3	39.3	39.3	39.2	0.11	0.693	0.473	0.282
Week 1	39.2[a]	39.1a	39.4[b]	39.4[b]	0.12	0.001	0.474	0.354
Week 2	39.0[a]	39.1[ab]	39.4[c]	39.2[b]	0.11	0.002	0.456	0.012
Week 3	38.8[a]	39.0[a]	39.5c	39.3[b]	0.12	0.000	0.891	0.022
Week 4	39.1[a]	39.1[a]	39.4b	39.4[b]	0.10	0.000	0.986	0.733
Respiration rate pre- and post-exercise								
Pre	64[a]	82[ab]	110[b]	72[ab]	17.3	0.131	0.400	0.018
Post	104[a]	114[ab]	142[b]	91[a]	14.9	0.433	0.048	0.004
Difference	40	32	32	19	17.5	0.423	0.396	0.849

[a, b, c] Means with different subscripts are different ($P<0.05$)

Figure 1 Mean respiration rate for C (◆), CElitox (■), T (●) and TElitox (▲) groups recorded from baseline and throughout the experimental period at 9pm. * P<0.05

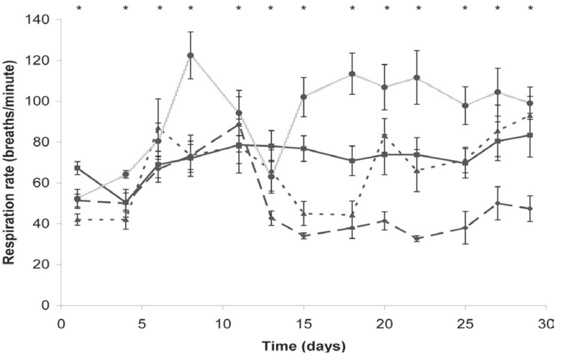
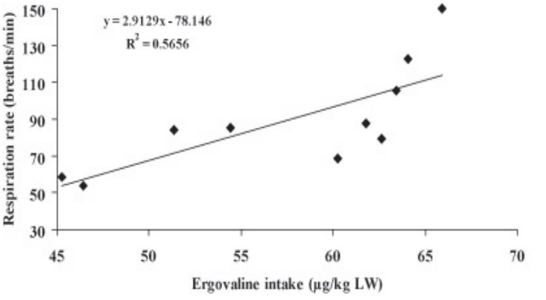


Figure 2 Relationship between respiration rate and ergovaline intake for T group in week 1 of the experiment.



fed T appeared to have moister faeces; mean faecal moisture measurements were not significantly different (Table 1).

For weeks 2-4, the T group had a higher respiration rate relative to others. Respiration was lower for TElitox relative to T but, reflecting baseline observations, the interaction was significant. On the control treatment respiration was increased by Elitox®.

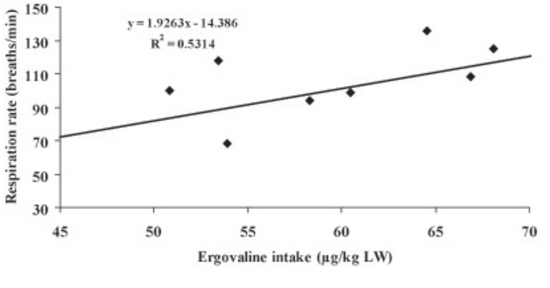
From week 1, morning and night observations both showed a gradual decline of respiration rate for C and an increase for T (Fig.1). While the pattern was similar, differences between T and other groups was greater at night. Regression analysis revealed positive relationships between respiration rate and intake of ergovaline for T (Figs. 2-3) and TElitox (data not shown). There were no significant differences in the changes to respiration rate and rectal temperature brought on by exercise. However, the post-exercise respiration rate was greater for T than TElitox; this Elitox® effect was not observed in sheep on the control diet.

Discussion

Our measurements of toxin intake exceeded the 30 μg/kg liveweight/day level at which Burke *et al*. (2006) reported effects on sheep. Lolitrem B intake was below the 100 μg/kg liveweight/day level that maybe required to induce staggers (McLeay & Smith 1999).

High environmental temperatures predispose animals suffering from endophyte toxicoses to greater physiological stress and production losses (Spiers *et al.* 2005; Bluett *et al*, 1999; Fletcher *et al.* 1994; Fletcher *et al*. 1999). In winter we were able to observe the effect of the toxin without the interaction of a high environmental temperature.

Figure 3 Relationship between respiration rate and ergovaline intake for T group in week 4 of the experiment.



Most studies show a decrease in liveweight gain due to wild endophyte (Fletcher *et al.* 1999, Burke *et al*. 2005) but effects can be variable (Kramer *et al.* 1999; Smith & Towers 2002). The lower intake on TElitox (especially in weeks 1-2) was possibly due to palatability of Elitox®; some individuals adjusted slowly to the diet change. Conversely, Munday-Finch & Garthwaite (1999) fed lolitrem B to mice and found intake decreased. This was attributed to palatability; mice were able to detect the toxin and, on return to a control diet, intake increased.

Intake results reflected individual variability. Animals vary also in ability to cope with toxicosis (Spiers *et al.,* 2005). Animal behaviour dynamics may have affected individuals' access to feed. Impatient animals were sometimes seen to kick and jump at the animal in the feeder. The low ADG on C was associated with some inclusion of fines in that diet late in the experiment. As intake decreased in some animals, the use of fines was stopped and intake increased to previous levels.

Variability was less marked for respiration rate and rectal temperature – especially for C groups. Elitox® may be effective in reducing endophyte-induced heat stress. TElitox ewes had a more variable respiration rate, however, it increased during treatment and was significantly lower than T and similar to C/CElitox. Over treatment, the relationship between ergovaline ingestion and respiration remained of similar strength for T but weakened for TElitox.

Rectal temperature decreased in C groups from the baseline which coincided with a decline in daily minimum temperature, while for T and TElitox average rectal temperature increased slightly. The T group maintained a steady high temperature whereas for TElitox temperature was lower in weeks 2-3. This difference was associated with a higher toxin intake by T. The increased core temperature induced by endophyte may assist animals to cope when the ambient temperature is low, possibly by stimulating metabolism.

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