The impact of endophyte on the health and productivity of sheep grazing ryegrass-based pastures

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Abstract

The health and production of sheep grazing perennial ryegrass with and without wild-type endophyte (*Neotyphodium lolii*) has been studied in several trials over a number of years. Lambs/hoggets grazing predominantly perennial ryegrass swards with endophyte developed moderate to severe ryegrass staggers in summer and autumn, while those grazing endophyte-free ryegrass did not. Lambs/hoggets grazing ryegrass with endophyte also had more dags, lower growth rates, lower plasma prolactin levels, higher body temperatures and respiration rates under warm humid conditions. Most of these adverse responses were more severe in summer and autumn when endophyte toxin concentrations were highest. Many of these symptoms are similar to those described for the "autumn ill thrift" syndrome in New Zealand.

Keywords: dags, endophyte, flystrike, growth rates, hyperthermia, *Neotyphodium*, perennial ryegrass, prolactin, ryegrass staggers, sheep

Introduction

The majority of New Zealand's high fertility sheep pastures are perennial ryegrass-based. The persistence and productivity of these ryegrass pastures are enhanced by the presence of the endophyte *Neotyphodium lolii* (Prestidge *et al.* 1982; Fletcher 1986). Most perennial ryegrass in New Zealand is infected with a wild-type strain of *N. lolii* which also makes the ryegrass toxic to grazing sheep and other livestock (Fletcher & Harvey 1981; Fletcher *et al.* 1990; Fletcher 1993; Fletcher & Easton, 1997).

The first probable recording of ryegrass/endophyte toxicity in New Zealand was ryegrass staggers in 1906, although it was not linked with endophyte at the time (Gilruth 1906). Research in the 1950s came close to establishing a link between animal health and ryegrass endophyte, however no evidence incriminating endophyte was discovered (Cunningham 1958; Cunningham & Hartley 1959; Lloyd 1959).

From 1960 through to 1980 and possibly later, loosely defined syndromes such as "summer/autumn ill thrift" in cattle and sheep were described and reported by farmers. This syndrome is characterised by negligible weight gains over variable periods in summer and autumn despite ample pasture, anthlemintic drenching and correction of known mineral deficiencies (Scott *et al.* 1976). Many sheep farmers would add dags, scouring and unthrifty appearance to the collection of symptoms. These symptoms are still common on many sheep farms in the summer autumn period throughout New Zealand but the term "summer" or "autumn ill thrift" is less often used by farmers.

With our current knowledge of animal responses to grazing perennial ryegrass pastures with endophyte over summer and autumn, it is apparent (retrospectively) that many of the individual and collective symptoms of the ill thrift syndrome are also characteristic of general endophyte toxicosis (Fletcher *et al.* 1996). The ill thrift syndrome was confined to summer and autumn when endophyte toxin levels are known to be highest. Endophyte effects on sheep health and production obviously had an impact on sheep grazing systems before 1981, when ryegrass staggers was first linked with endophyte, and probably from the time ryegrass was introduced into New Zealand (Fletcher & Harvey 1981; Fletcher *et al.* 1996). Sheep grazing endophyte-free ryegrass in trials conducted over the last 18 years have shown none of the symptoms associated with "summer/autumn ill thrift" when available forage was not a limiting factor.

While close parallels can be drawn between "summer" or "autumn ill thrift" and endophyte toxicosis it must be acknowledged that many factors other than endophyte and pasture quantity could, and do, impact on sheep health and performance in summer and autumn throughout New Zealand. However the summer/autumn period is still identified by farmers as one of the most difficult periods for maintaining good health and performance in grazing sheep. Most other critical periods during the year can be accounted for by lack of feed or the inability to match pasture growth to animal demands.

The known health and production problems in sheep associated with grazing perennial ryegrass with its endophyte now include:

- reduced growth rates, or even weight loss
- ryegrass staggers
- increased dags and flystrike

- increased heat stress
- reduced plasma prolactin levels.

This paper reviews research in New Zealand determining the impact of wild-type ryegrass endophyte on sheep.

Methodology

Data presented in Tables 1 and 2 are from hoggets (spring) and weaned lambs (summer/autumn) grazing endophyte infected and endophyte-free perennial ryegrass in a number of trials at AgResearch, Lincoln from 1992. Trial (a) was conducted on Templeton silt loam soil without irrigation while trials (b-e) were on irrigated Wakanui silt loam soils. Trial design and methodology were basically the same for all trials.

Three replicates of perennial ryegrass (no clover) cv Grasslands Nui with or without endophyte were grazed for minimum periods of 30 days in spring, summer and autumn with a minimum of 10 animals/0.15 ha replicate paddock (30 per treatment). Measurements included: liveweight change (gm/day), ryegrass staggers score (0–5 ascending scale), dag score (0–5 ascending scale), flystrike (% of flock struck over grazing period), plasma prolactin (ng/ml), rectal temperature (°C), respiration rate (breaths/minute) (Fletcher 1999).

Table 1 Effect of endophyte in Nui ryegrass on responses of grazing hoggets in spring.

Response (Means)	Trial	Wild-type	Nil	Lambs/hoggets per treatment	Significance[†]
Liveweight gain (gm/hd/day)				**no.**	
	a	115	167	36	*
	b	99	145	120	*
	c	165	191	120	*
Dags (0–5 scale)					
	a	1.6	0.4	36	*
	b	1.4	0.6	120	**
Rectal temperature (°C)					
	b	40.3	40.0	120	NS
	c	40.3	40.0	120	*
	d	39.9	39.8	120	NS
Respiration rates (Breaths/min)					
	b	95	86	36	NS
	c	93	83	120	NS
	d	76	69	120	NS
Prolactin (ng/ml)					
	a	105	152	36	*
	b	101	333	120	**

[†]** p<0.01; * p<0.05; NS not significant.

Trial dates and duration
(a) Spring 1992–Autumn 1995 (b) Autumn 1994–Autumn 1996 (c) Autumn 1996–Autumn 1998 (d) Autumn 1998–Autumn 1999 (e) Spring 1998–ongoing

The effect of endophyte on body temperature and respiration rate becomes evident when the animal's thermo-regulatory system is challenged. To ensure the animal's thermo-regulatory system was challenged, lambs or hoggets on trials c and d (Tables 1 and 2) were removed from their pastures and housed for three hours in a room where temperature and humidity were increased to >30°C and 70% relative humidity. Rectal temperatures, respiration rates and prolactin levels where then measured under these conditions. On trials a & b these measurements were taken on hot days, in the field. Respiration rate and rectal temperatures of healthy resting lambs/hoggets, on a cool day (12–15°C) are normally in the range of 20–30 breaths per minute and 39.6–39.9°C respectively.

Liveweight change

Liveweight change is one of the single most important parameters for evaluating a grass or pasture for sheep production. In the case of ryegrass/endophyte associations it encompasses most of the toxic responses in sheep to some degree (Fletcher & Easton 1997).

Reduced growth rates in lambs and hoggets grazing ryegrass with wild-type endophyte (pure stands) have been reported (Fletcher 1983; Fletcher & Barrell 1984; Fletcher & Sutherland 1993a; Fletcher *et al.* 1996; Fletcher & Easton 1997; Fletcher 1999). A summary of endophyte effects, including growth rates, from a number of other, unpublished, lamb/hogget grazing trials is presented in (Tables 1 & 2). Spring growth rates of lambs and hoggets can be depressed by endophyte in perennial ryegrass but to a lesser degree than in summer and autumn when toxin levels are highest (Fletcher 1999). Watson *et al.* (1999) have also recorded lower growth rates in lambs grazing endophyte infected compared to endophyte-free ryegrass pastures.

There can be considerable year to year variation and lambs and hoggets are more severely affected than ewes. Effects may also vary from region to region, with little effect of endophyte on lamb growth rates documented in the cool moist Southland region (Eerens *et al.* 1997). Studies with different levels of endophyte infection in ryegrass pastures indicate a linear reduction in growth rate of 3–4gm/day for every 10% increase in endophyte infection in spring and up to 10gm/day in summer/autumn.

Variable clover content in pastures has been identified as a possible confounding factor in several

Table 2 Effect of wild-type endophyte in Nui ryegrass on responses of grazing hoggets/lambs in summer/autumn.

Response (Means)	Trial	Wild-type	Nil	Lambs/hoggets per treatment	Significance
Liveweight gain				no.	
(gm/hd/day)					
	a	30	52	108	**
	b	57	87	72	*
	c	23	120	60	**
	d	15	200	60	**
	e	80	100	30	*
Ryegrass staggers					
(0–5 scale)					
	a	3.3	0	108	**
	b	2.5	0	72	**
	c	3.2	0	60	**
	d	3.5	0	60	**
Dags					
(0–5 scale)					
	a	2.3	0.3	108	**
	b	1.7	0.8	72	*
	c	0.6	0.2	60	*
Flystrike					
(% struck)					
	a	15	2	108	**
	b	30	2	72	**
Rectal temperature					
(°C)	a	40.5	40.2	108	*
	b	40.6	40.3	72	*
	c	40.5	40.0	60	**
	d	40.7	40.4	60	*
Respiration rate					
(Breaths/min)					
	b	84	64	72	*
	c	97	73	60	*
	d	115	83	60	**
Prolactin					
(ng/ml)					
	a	90	198	108	**
	b	5	27	72	**
	c	96	185	60	*

** p<0.01; * p<0.05; NS not significant.

Trial dates and duration
(a) Spring 1992–Autumn 1995 (b) Autumn 1994–Autumn 1996 (c) Autumn 1996–Autumn 1998 (d) Autumn 1998–Autumn 1999 (e) Spring 1998–ongoing

trials where no significant effect of wild-type endophyte on animal performance was found (Eerens *et al.* 1992; Cosgrove *et al.* 1993; Thom *et al.* 1994).

Intake is one of the major determinants of liveweight change (Ulyatt 1973). However no difference in intake was found between lambs grazing ryegrass with wild-type endophyte and those grazing the same ryegrass without endophyte over three years (Fletcher unpublished data). There was, however, significant difference in liveweight change between the lambs grazing these two treatments. While there was no difference in intake measured, in some instances greater herbage residuals on ryegrass with wild-type endophyte than on endophyte-free ryegrass, suggests an intake difference or higher herbage growth rates in endophyte infected ryegrass.

Ryegrass staggers

One of the most obvious impacts of wild-type endophyte in ryegrass, on sheep, is the neuromuscular disorder ryegrass staggers (Fletcher & Harvey 1981; Fletcher 1982; Fletcher *et al.* 1990; Fletcher *et al.* 1991; Fletcher *et al.* 1993; Fletcher *et al.* 1996; Fletcher & Easton 1997).

The symptoms and development of ryegrass staggers have been well described (Keogh 1973). It is caused mainly by the tremorgenic endophyte toxin, lolitrem B, (Gallagher *et al.* 1981) although other endophyte-related tremorgens may have a minor influence.

A scoring system on a 0–5 ascending scale has been developed to measure the seriousness of ryegrass staggers in individual animals (Keogh 1973). In its early stages ryegrass staggers is characterised by slight tremors in the neck and lower jaw, after enforced exercise, detectable only by palpation. As the disorder develops animals will begin to stagger and fall after enforced exercise and may become progressively worse until they fall at the slightest disturbance (Keogh 1973). They may remain down for several hours or be unable to regain their feet at all. Animals in this condition will obviously die if left unattended and/or if they are not removed from the toxic pasture.

Environmental and pasture conditions conducive to ryegrass staggers have been described (Keogh 1973). However, serious ryegrass staggers has recently become more common on rank green leafy ryegrass–white clover pastures (Fletcher 1998). It is normally confined to the late December to late April period although there have been recent reports of serious outbreaks as early as November and as late as June. The risk of ryegrass staggers in sheep grazing perennial ryegrass with wild-type endophyte is always present if there is a sufficient concentration of lolitrem B (>2 ppm) in pasture and the animal ingests enough of it (di Menna *et al.* 1992; Fletcher *et al.* 1996).

Lolitrem B, the main toxin causing ryegrass staggers, appears to be relatively stable and as such concentrations in hay or threshed ryegrass straw, made from endophyte infected ryegrass, can remain high. Animals consuming this straw can develop moderate to severe ryegrass staggers. Ergovaline levels also remain high in straw, thus maintaining the potential for toxicity associated with this alkaloid also.

While both lolitrem B and ergovaline have been detected in silage, their stability in silage and the toxicity of the silage to animals has not been demonstrated.

The most obvious effects of ryegrass staggers on the sheep grazing system are the constraints it places on "normal" stock management. Depending on the time and duration of the outbreak, ryegrass staggers may interfere with day to day stock movement, dipping, drenching, shearing, dagging (crutching), drafting and transport. While the economic impact of such disruptions to sheep management is less tangible, there is clearly a cost involved, in extra time to move stock and losses due to delays in preventative operations. In its advanced stages ryegrass staggers will obviously impair the animal's ability to graze effectively, reducing intake and consequent growth rates. However growth rates are affected by endophyte even in the absence of staggers (Fletcher & Barrell 1984; Fletcher unpublished data).

Ryegrass staggers often occurs at or around mating in many regions but there is no evidence that this has any direct effect on subsequent reproductive performance or lamb numbers. If alternative "safe" non-toxic pastures are unavailable, when ryegrass pastures are toxic, then the management options for reducing ryegrass staggers are very limited and confined to supplementary feeding.

Many of the costs of ryegrass staggers *per se*, to the sheep farmer, are difficult to quantify. Losses due to deaths and the cost of any feed supplement are tangible, but costs of extra time to move stock and losses due to delays in preventative operations are more difficult to establish.

Dags and flystrike

The effect of endophyte in ryegrass on faecal moisture, dags and flystrike has been documented but is not as well recognised or publicised. Although considerable individual animal and seasonal variation exists, sheep grazing perennial ryegrass with endophyte are more likely to develop dags (Eerens *et al.* 1992; Pownall *et al.* 1993; Fletcher 1993; Fletcher 1999; Fletcher unpublished data) (Tables 1 and 2). As a consequence, these sheep are more susceptible to flystrike (Table 2). While the basic cause of dags and the mechanisms involved are not well understood, sheep grazing ryegrass with endophyte have higher faecal moistures than those grazing the same ryegrass without endophyte (Pownall *et al.* 1993; Fletcher *et al.* 1993; Fletcher unpublished data). High faecal moisture has been identified as a contributing factor in the formation of dags (Waghorn *et al.* 1999).

The ryegrass/endophyte metabolite responsible for dags and flystrike has not been identified. Peramine does not appear to be implicated, since sheep fed peramine alone in a lucerne chaff diet did not have elevated faecal moisture or develop dags (Pownall *et al.* 1995). Sheep grazing a ryegrass/endophyte association which does not produce lolitrem B, but still produced ergovaline, had similar prevalence and severity of dags to those grazing a wild-type association (producing lolitrem B) (Fletcher unpublished data).

From a grazing perspective, this suggests lolitrem B, in endophyte infected ryegrass, is not a major cause of dags in sheep. However extracts of lolitrem B and other tremorgens have been shown to have a significant affect on smooth muscle and gut motility when administered to sheep (Smith *et al.* 1997; McLeay *et al.* 1999). Although the characteristics of ergopeptine alkaloids suggest they have the potential to influence faecal moisture, the possibility of other yet unidentified ryegrass/endophyte metabolites, either acting alone or in synergy, must also be considered.

Dags or scouring are often used by farmers as indicators of the need to drench with anthelminthics (Fletcher *et al.* 1996). Anthelmintics however will have no effect on scouring and dags caused by ingestion of perennial ryegrass with endophyte.

Besides being an initiating factor in flystrike, dags are unsightly, cause staining in fleece wool during yarding and create contamination problems at slaughter and processing.

Flystrike alone is estimated to cost the New Zealand sheep industry $37 million annually (Heath & Bishop 1995). However there is currently little that can be done to prevent dags and flystrike associated with endophyte, other than crutching and/or dipping or grazing sheep on non-toxic pastures. Identifying the toxins responsible for increased dags and the mechanisms involved should be an objective for future research.

Hyperthermia

Ergovaline, produced by ryegrass with wild-type endophyte, constricts the blood vessels and capillary vessels in the skin and peripheries of animals. This is well recognised in cattle grazing E+ fescue, which also produces ergovaline and causes hyperthermia (Schmidt & Osborne 1993). It is also believed to be responsible for the elevated temperatures and respiration rates reported in sheep grazing ryegrass with wild-type endophyte under warm humid conditions (Fletcher 1993; Fletcher & Easton 1997; Fletcher 1999) (Tables 1 and 2). Heat stress or thermo-regulatory dysfunction is best measured through body temperature and respiration rate. Increased respiration rate or panting is a normal thermo-regulatory response to reduce heat load. While the symptoms of hyperthermia are occasionally observed in sheep in the field, it is likely to be more common at a sub clinical level. The impact of endophyte related heat stress on sheep production systems will depend on climate, current ambient conditions and of course the amount of ergovaline ingested. It is likely to be especially prevalent in the warm humid conditions of northern New Zealand.

Plasma prolactin

Reduced plasma prolactin levels in sheep are one of the most consistent responses to grazing ryegrass with wild-type endophyte especially in hot weather. However the impact of this response on overall health and production of sheep is not well understood (Fletcher & Easton 1997). Prolactin concentrations are higher in sheep grazing endophyte-free ryegrass in spring and the difference normally increases in summer and autumn (Tables 1 and 2).

Plasma prolactin has been used in trials to indicate ergovaline ingestion in sheep and other grazing animals. There is no evidence that peramine, lolitrem B or other tremorgens affect prolactin levels. However, prolactin may itself be a mediating factor in some adverse animal responses to endophyte, such as increased faecal moisture, dags and heat stress.

While links with mammary development, lactation, fluid balance, appetite, thermo-regulation, wool growth and immune function have been documented or proposed in a range of species, the action of plasma prolactin appears to vary considerably between species (Turkington & Frantz 1972; Nicol & Bryant 1972; Cincotta & Meir 1985; Houdebine *et al.* 1985; Faichney & Barry 1986; Hiestand & Mekler 1986; Gloria *et al.* 1994).

A "normal" or base level of prolactin for sheep has not been established because of large individual animal variation and the number of external environmental and animal factors that also influence levels. These include nutrition, climatic conditions, seasonal and daily rhythms, stress and physiological state of the animal. Therefore in the absence of samples from sheep grazing endophyte-free controls, prolactin *per se* is currently of little value as an indicator of endophyte effects.

A database of prolactin concentrations, measured under a range of conditions throughout the year, is being developed to determine broad baseline levels, under specific conditions. The roles and impact of prolactin in sheep physiology, especially in responses associated with endophyte, warrant further investigation.

Reproduction

Apart from a delay in lambing date there are no documented effects of ryegrass endophyte affecting reproductive performance in sheep (Eerens *et al.* 1997; Watson *et al.* 1999). This delay in lambing may be the result of a delay in onset of ovulation in ewes or a prolonged gestation. While it has been proposed that endophyte may lower testosterone levels in grazing rams there is no documented evidence to support this. However, further studies on effect of endophyte on reproduction performance of rams are warranted.

Small flocks in relatively small paddocks with a low ewe to ram ratio, even in systems trials, present an artificial environment for reproductive studies. These are far removed from a "normal" on farm situation. Future studies on effect of endophyte on sheep reproduction need to be conducted with large trial flocks (>200 ewes) and ram to ewe ratios typical of commercial farming.

Management and the impact of endophyte

In a total sheep farming system, the overall impact of endophyte effects may be less than trial results indicate or be considerably modified by a number of farm practices or management decisions. Apart from seed crops, ryegrass is usually sown with a legume or as part of a pasture mix. Any non-toxic companion grass or legume species will dilute the impact of the endophyte toxins to varying degrees.

Moisture deficit and increased available soil nitrogen both increase ryegrass/endophyte alkaloids, thus increasing toxicity (Lane *et al.* 1997). The impact of these will therefore vary with location and management.

Farms that manage ewes to lamb early, draft most lambs before Christmas, buy in ewe replacements and have good spring and early summer growth may expose relatively small numbers of young growing animals to the peak of endophyte toxins.

The impact of such a system will be less than that on a farm carrying a large proportion of weaned lambs and ewe hoggets through the summer/autumn period.

Summary

Under controlled trial situations, the health and production of sheep grazing perennial ryegrass in summer and autumn can be severely affected by endophyte. The translation of these effects to farm systems may be influenced by many factors. These include the farm type, location, balance of toxic and non-toxic pastures in the system, the number and age of sheep exposed to peak toxin levels, climatic conditions and management.

The development and potential release of non-toxic endophytes in ryegrass which are persistent should enable farmers to capitalise on the benefits of endophytes while reducing or eliminating animal toxicosis (Fletcher

1999). However there are areas of endophyte infected ryegrass which are unlikely to ever be replaced because of topography. The best that can be expected for such pastures is a prophylaxis or cure for ryegrass staggers. In areas where endophyte toxicity occurs, there are often few management options to prevent or alleviate the problem, apart from moving stock to non-toxic pastures. Most adverse effects on livestock are likely to persist as long as the endophyte and associated toxins remain in the pastures.

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