PLANT RESISTANCE TO PESTS

EFFECT OF PERENNIAL RYEGRASS CULTIVAR AND SOWING RATE ON NEMATODE PASTURE PEST ABUNDANCE

Lee T. Aalders[1*], Nigel L. Bell[1], Mark M. McNeill[2], Derrick J. Wilson[1] and Julia Lee[3]

[1]AgResearch Ltd, Hamilton, New Zealand

[2]AgResearch Ltd, Lincoln, New Zealand

[3]DairyNZ, Hamilton, New Zealand

Corresponding author:
lee.aalders@agresearch.co.nz

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Introduction

New Zealand dairying is largely based on a perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*) pasture mix (Easton et al., 2011). In recent years farmers have raised concerns that there is a lack of persistence with newly sown pastures (Tozer et al., 2011). A five year project investigating pasture persistence has being carried out at two farm locations in New Zealand: Waikato (North Island) and Canterbury (South Island). This was set up to compare different perennial ryegrass cultivars and sowing rates at different locations. Tetraploid and diploid perennial ryegrass (*Lolium perenne*) cultivars were sown at five rates (6–30 kg/ha) in an attempt to define optimal sowing rates. White clover was sown along with the ryegrass. The effect on soil borne plant parasitic nematodes populations of a tetraploid and a diploid perennial ryegrass cultivar at the low (6 kg/ha) and the high (30 kg/ha) sowing rates were examined.

Methods

The two dairy farm sites were sown in autumn 2011. Four perennial ryegrass cultivars were sown in plots at five nominal seeding rate treatments (6, 12, 18, 24 and 30 kg seed/ha). Main treatment plots (cultivar, 540 m^2) were divided into sub-plots (seeding rate, 108 m^2), arranged in a randomised split-plot design with five replicates. All cultivars were infected with the fungal endophyte *Epichloë festucae* var. *lolii* (formerly *Neotyphodium lolii*). All plots were over-sown with 8 kg/ha of Superstrike®-coated Tribute white clover seed (*Trifolium repens*; equivalent to about 5 kg/ha of bare seed) (J. Lee, pers. comm.). Two of the four cultivars, one diploid and one tetraploid, were sampled annually for pasture pests. Sampling of the unirrigated Waikato site occurred in autumn, while the irrigated Canterbury site was sampled in spring 2012 and 2013, and autumn 2014 and 2015.

Nematodes were sampled by taking ten 2.5 cm (dia.) × 10 cm (deep) cores from within each plot. The cores from each plot were bulked then crumbled and mixed by hand. A 100 g subsample of fresh soil was placed in a modified Whitehead tray and nematodes were extracted using the method of Bell and Watson (2001). Briefly, each sample was placed on a two-ply paper tissue, supported by two layers of nylon gauze within a shallow tray to which 500 ml of tap water was added. The tray was left for 72 hours, after which the liquid was poured into a 1 L plastic beaker, left to settle for 4 hours, then gently reduced to 100 ml volume by removing the supernatant. The 100 ml

samples were transferred to 100 ml plastic beakers and allowed to settle for 4 hours before reduction to a final volume of 20 ml. The total number of nematodes and plant parasitic nematodes were counted using a Doncaster dish (Doncaster, 1962) under a stereo-microscope at 40–80× magnification; plant parasitic nematodes were identified to genera. To allow for valid comparison across treatments, results are presented as nematodes per 100 g of dry soil. This was determined from the number of nematodes in the sample divided by (100/soil fresh weight)*(soil dry weight). The total number of nematodes (plant parasitic, bacterial and fungal feeding species) from each plot was recorded. The plant parasitic nematodes were further identified and counted to six genera (in approximate order of most to least damaging: root knot (*Meloidogyne*); cyst (*Heterodera*); lesion (*Pratylenchus*); pin (*Paratylenchus*); spiral (*Helicotylenchus*) and stunt (*Tylenchorhynchus*). Species identification of the three most damaging genera was determined using primers that targeted different regions of rDNA and sequences compared to sequences of the NCBI database. Three to four specimens of *Meloidogyne*, *Heterodera* and *Pratylenchus* were sequenced from each site.

Results and Discussion

More cyst nematodes (*Heterodera* sp.) were found beneath the low than the high sowing rate treatment for most years at both sites and this difference was significant when pooled across all years and ryegrass cultivars (Figures 1 and 2). *Heterodera* were sequenced as belonging to the *H. schachtii* group from both sites (this group includes clover feeding species), while the Canterbury site also had nematodes that were identified to the *H. avenae* group, grass feeding species. This greater abundance associated with the lower sowing rate is most likely a consequence of greater clover abundance in those plots.

Figure 1.
Number of cyst nematodes (natural log scale) at the Waikato site. Error bar is LSD 5%.

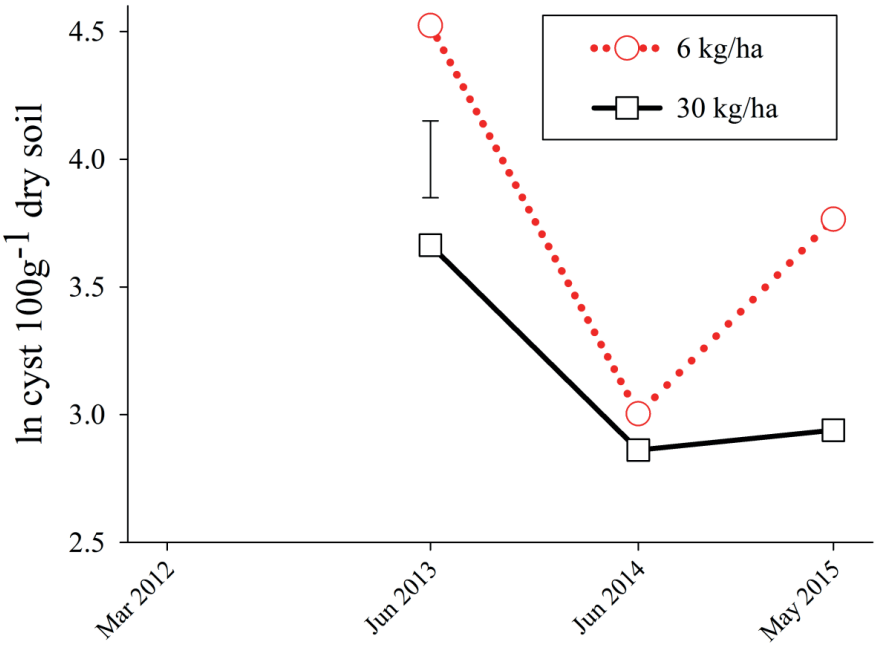
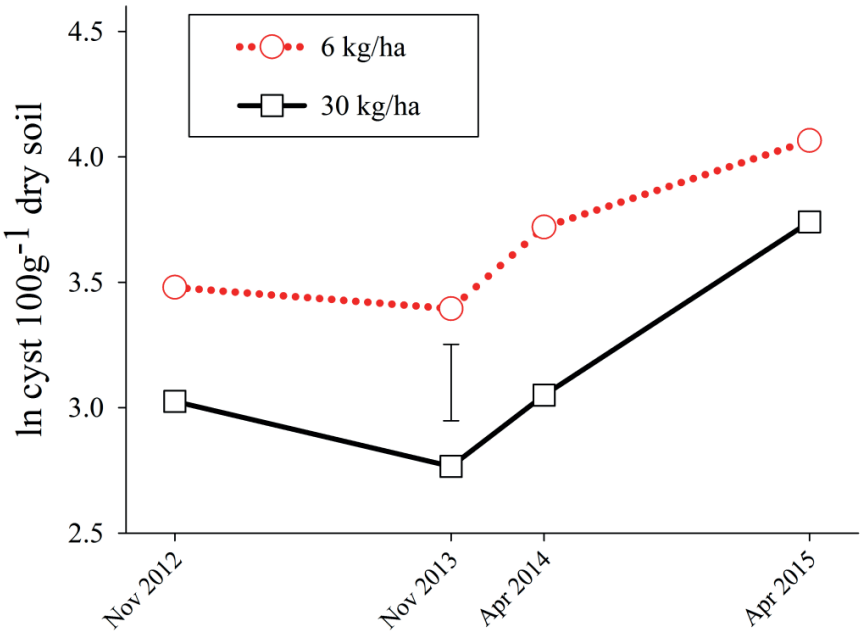


Figure 2.
Number of cyst nematodes (natural log scale) at the Canterbury site. Error bar is LSD 5%.



There were consistently more lesion nematodes (*Pratylenchus* sp.) beneath the tetraploid than the diploid cultivar, at both sites, and this was significant when pooled across all years and both sowing rates (Figures 3 and 4).

Figure 3.
Number of lesion nematodes (natural log scale) at the Waikato site. Error bar is LSD 5%.

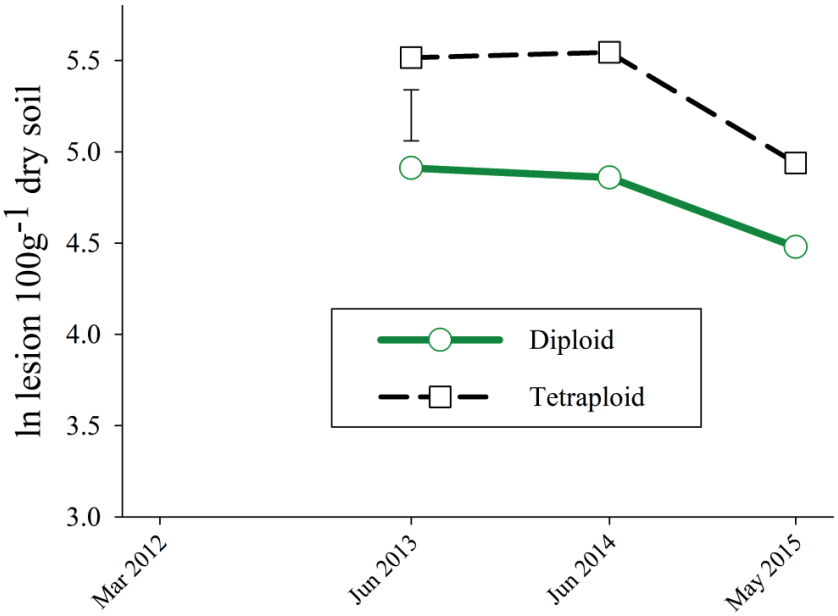
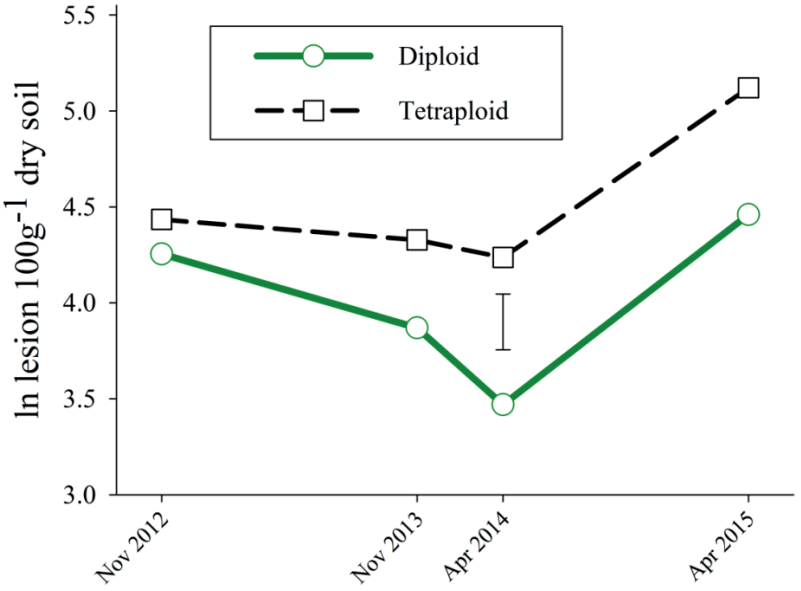


Figure 4.
Number of lesion nematodes (natural log scale) at the Canterbury site. Error bar is LSD 5%.



Pratylenchus crenatus was identified from both sites; a second *Pratylenchus* species was found at the Canterbury site but is as yet unidentified**.** *Pratylenchus crenatus* is extremely polyphagous with a host range that includes both grasses and clovers (Willis et al., 1982; Knight et al., 1997; Simard et al., 2008; Yu, 2008; Mekete et al., 2011; Esteves et al., 2014). It is not known what was causing the significantly greater abundance of these nematodes beneath the tetraploid cultivar; it may be due to: more total root mass beneath tetraploid plots (including grass, clover and other plants) or more ryegrass root matter beneath tetraploid plots; or a preference for roots of this cultivar by the nematode.

There was no consistent effect of either ryegrass cultivar or ryegrass sowing rate on root knot (*Meloidogyne* sp.), pin (*Paratylenchus* sp.), spiral (*Helicotylenchus*) or total nematode abundance.

Ryegrass cultivar and sowing rate can influence the abundance and diversity of plant parasitic nematodes present in the soil. These nematodes are an additional burden on plant resources, so taking this burden into account will help in understanding the trajectory of pasture persistence post-sowing.

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