

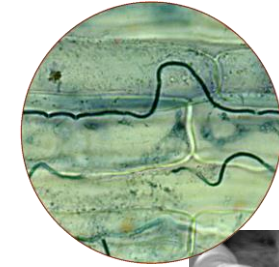
Stability and Purity of *Epichloë* Endophyte Infection in New Zealand Ryegrass Pastures

David Hume and co-authors from AgResearch
New Zealand

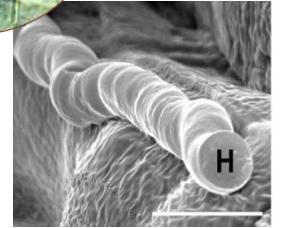




Perennial ryegrass
(*Lolium perenne*)



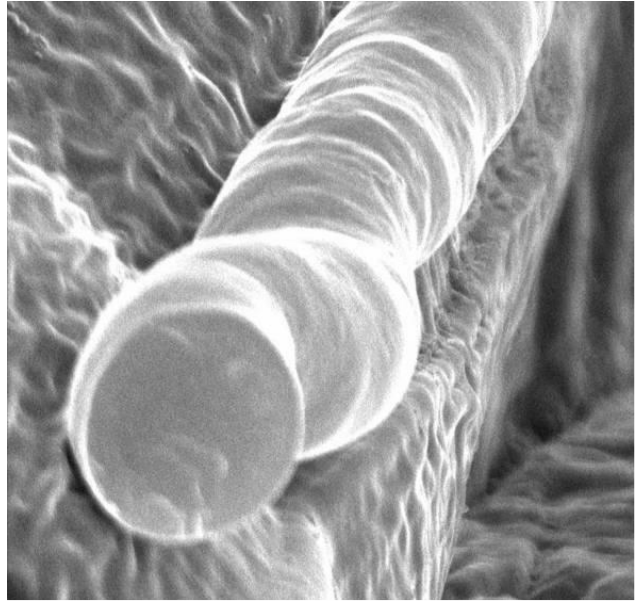
Epichloë fungal
endophyte



- **Perennial ryegrass** is the most widely sown grass in pastoral farming
- **Typically infected with fungal endophyte** (*Epichloë festucae* var. *lolii*), as this mutualist fungal endophyte assists the plant in resisting biotic and abiotic stresses
- **Elite proprietary ryegrass cultivars infected with selected strains of endophytes** have become widely used - now 90% of seed sales!
- **Selected endophytes displaced the use of the Standard (wild-type) mammalian-toxic strain of endophyte in sown ryegrasses, as the Standard strain was known to cause significant financial losses** to livestock farmers
 - Production losses (9% less milk solids), ryegrass staggers, heat stress
- **Selected endophyte strains**
 - **reduce or eliminate toxicity** to livestock
 - while **maintaining** to varying degrees **the positive agronomic benefits** when the grass host is subject to stresses, particularly under invertebrate pest pressure



What is *Epichloë* fungal endophyte?



Grass-*Epichloë*
fungal endophyte
associations are:

- Mutualistic
- Obligate
- Asymptomatic

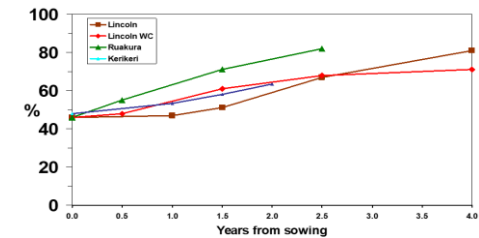
Fungal **endophyte** + Grass plant (internal view)

Endo = within - **Phyte** = plant



Background to this study

- **Significant benefits** for farmers if they use selected endophytes
- Ryegrass-endophyte associations have been intensively monitored in experimental trials; **little has been measured and reported from pastures grazed in commercial settings**
- **Pastures may become contaminated with non-sown endophyte** e.g. via hay feeding, seed in dung, buried seed, natural reseeding, incomplete kill of old pasture, sown seed
 - **Standard endophyte can outcompete selected strains** e.g. in a 50:50 mix with the AR1 strain, Standard endophyte increased 10 percentage points per annum (small plots)



- Concerns have been raised as to whether
 1. **Seed supply chain and the pasture renewal process have been able to deliver ryegrass pastures that are highly infected with selected endophytes**
 2. **Will Standard endophyte re-invade pastures and undermine the benefits of the selected endophytes?** as the ryegrasses that have naturalised in New Zealand are highly infected with the Standard endophyte



Dairy pastures sampled

North Island

- 13 pastures annually for 6 yrs (pastures 6-11 yrs old at final sampling)
- 5 ryegrass cultivars
- 3 sown endophytes: AR1, AR37, NEA2-brand (a blend of NEA2 and NEA6)



South Island

- 10 pastures annually for 6 yrs post-sowing
- 2 ryegrass cultivars
- 1 sown endophyte: AR37



New Zealand

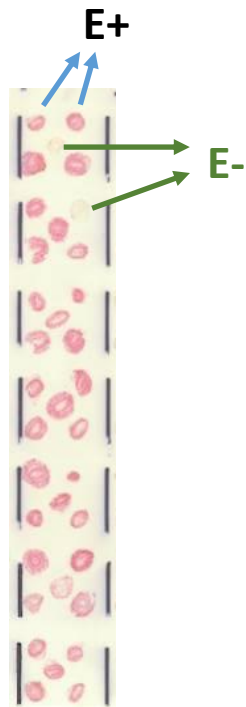
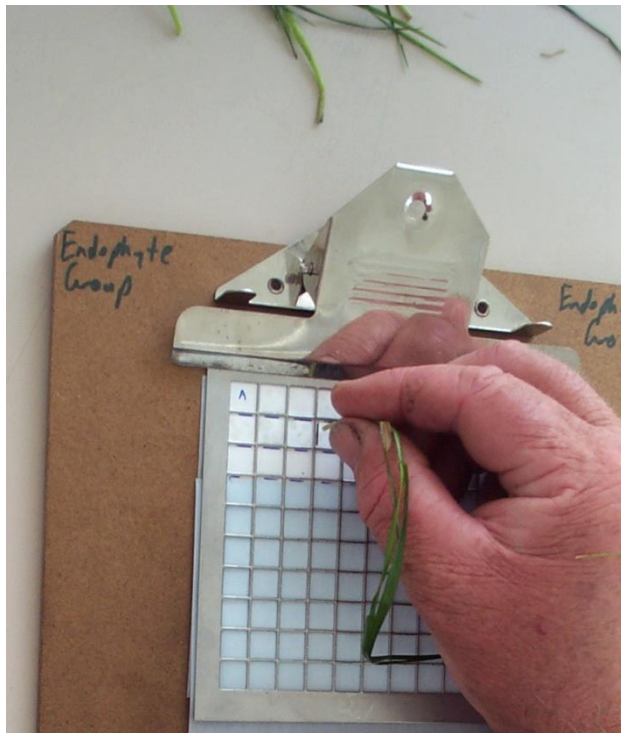




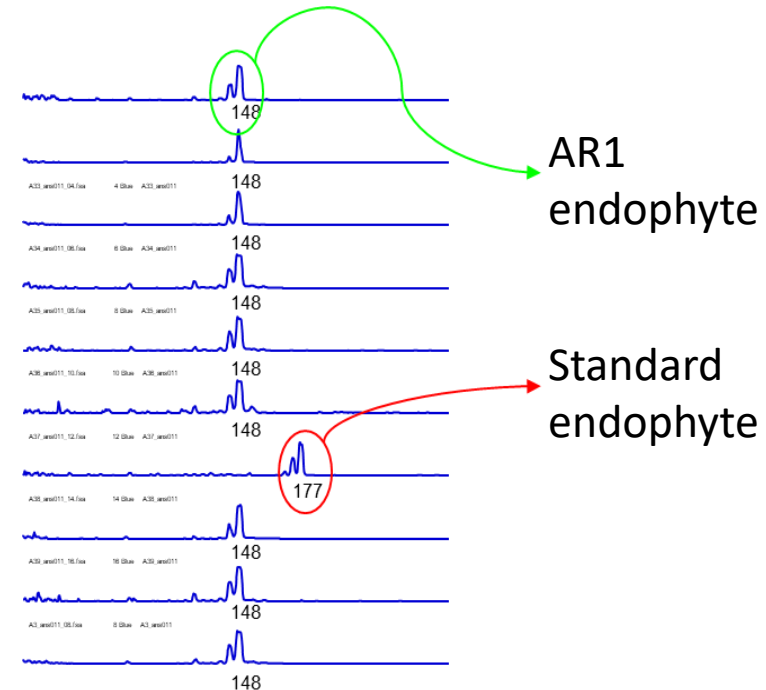
Ryegrass tillers tested for

- **Frequency of endophyte infection** by immuno-blot assay – 50-100 tillers per sample
- **Endophyte strain** determined by DNA (SSR) marker tests – a subset of 40 tillers per sample

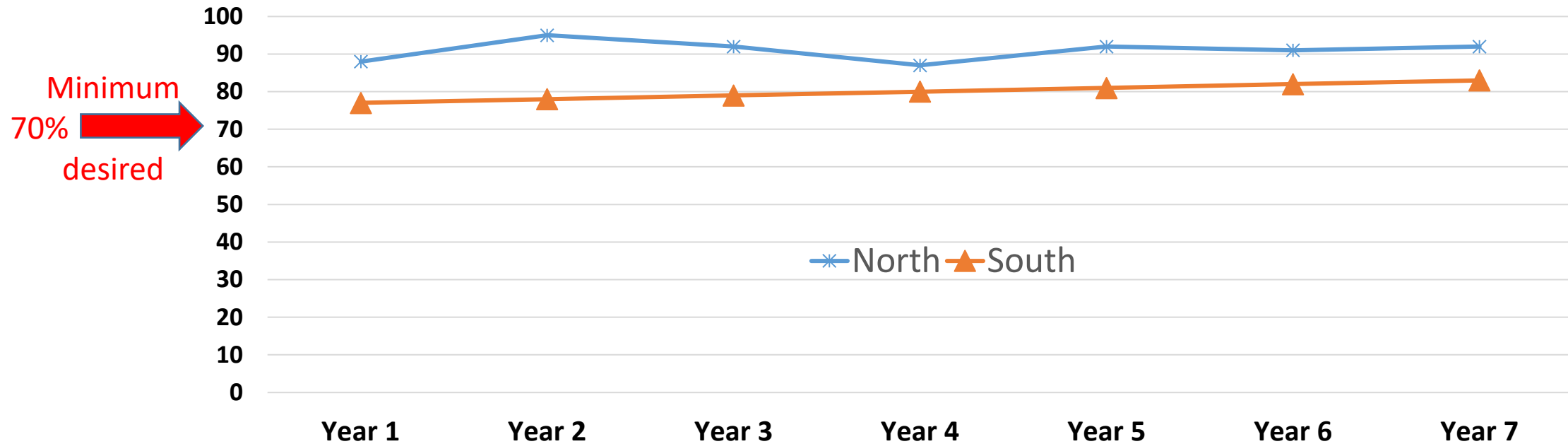
Immuno-blot



DNA test



% Endophyte infected tillers



- High rates of infection (means: 91% North, 80% South)
- Over time small increases (mean increases: 4% North, 6% South)
- South Island
 - Group 1 = 87% mean
 - Group 2 = 68% mean. Two pastures <70% - one due to incomplete kill of previous pasture (organic farm)

Note: 70% is the minimum needed to protect pastures from significant insect damage

Non-sown endophytes ('contaminating' endophytes)

	North Island	South Island	
Total non-sown endophytes - increase over time?	8% Yes: 3.2 → 10.3%	2.4% No	Overall, non-sown endophytes were low
Standard endophyte only - increase over time?	6.7% Yes: 2.8 → 9.2%	1.5% Yes: 0.4 → 2.8%	Mainly Standard endophyte, which increased over time

North Island - Standard endophyte

- 10 pastures: 5% mean, no change over time
- 3 pastures: 39% mean, increased over time 19 → 55% (1 pasture under-sown with uncertified seed)

South Island organic farm with incomplete pasture kill had 25% non-sown AR1 tillers

Additional North Island pasture

- Sampled from years 4 to 11 years post-sowing
- Inadvertently sown in a low viability seed line (NEA2/6 endophyte)
- 24 → 52% infected tillers (4% per annum)
- Increase mostly due to Standard endophyte (9 → 23%)
- Ratio of NEA2 to NEA6 (83:17) remained unchanged



Conclusions

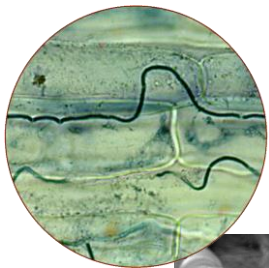
✓ In general

- the industry has been successfully delivering ryegrasses infected with selected endophytes on to dairy farms
- over time, selected endophytes maintained a high frequency of infected ryegrass tillers
- A minority of the pastures (2 out of 30) achieved less than the desired 70% infection minimum
- Major non-sown endophyte was Standard – as expected, due to the widespread nature of ryegrass infected with Standard endophyte, and in some pastures this increased over time
 - Of no real concern in the South Island where contamination was very low
 - However, in 3 of 13 North Island pastures, contamination increased over time to be on average over half of the tillers in a pasture. At this level, Standard endophyte is most likely to be reducing the animal production and health benefits of the sown selected endophytes
- In 2 cases, the management practices could have contributed to the poor outcome but otherwise the drivers for the low infection or increases in Standard endophyte could not be identified

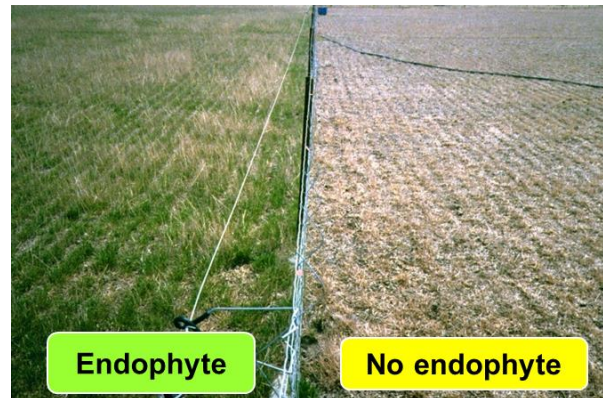
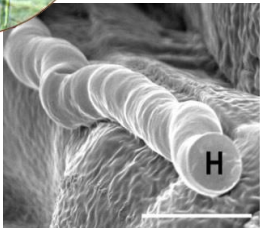
Want to know more about *Epichloë* endophytes in ryegrass and tall fescue?

Come to Thursday's 3:15 pm Thematic Session:

A model to reduce & eliminate endophyte toxicities in temperate pastures



Epichloë fungal endophyte



Common Toxic endophyte



Endophyte-free or non-toxic selected endophyte