Viability of endophytic fungus in different perennial ryegrass (*Lolium perenne*) varieties kept in different storage conditions

---

A thesis presented in partial fulfilment of the requirements for the degree of Master of Science

at

Massey University, Manawatū, New Zealand

Elizabeth Rose Kitson

2017

Abstract of a thesis submitted in partial fulfilment of the requirements for the degree of Master of Science.

Viability of endophytic fungus in different perennial ryegrass (*Lolium perenne*) varieties kept in different storage conditions

by

Elizabeth Rose Kitson

Epichloë endophytes form symbiotic relationships with cool-season grasses of the Pooideae family and are known to synthesise a range of bio-protective alkaloids. These alkaloids can provide the grass host with benefits for greater survival including; deterrence of herbivorous pests, increased persistence, better livestock health and protection from abiotic stressors. The commercialisation of novel endophytes is on the increase, and it is important to ensure the survival of the endophyte is maintained so their benefits can be realised.

This study examined the effects of different storage conditions on the viability of three commercial novel endophytes (AR1, AR37 and NEA2/6) and one pre-commercial novel endophyte (815). The different storage conditions were the top of a warehouse, the bottom of a warehouse and a temperature and humidity controlled cool store to simulate current commercial seed storage environments.

The viability of different endophytes decreases independently of grass seed germination ($p = NS$) however there are many factors influencing the endophyte survival. Over the one year storage period there were significant interactions between endophyte x ploidy (host), endophyte x location and endophyte x ploidy (host) x location. The pre-commercial endophyte, 815, had the largest reduction in viable endophyte when stored outside of the controlled cool store dropping 70 percentage points at the top of the warehouse, compared with AR37 (12 percentage points), AR1 (16 percentage points), and NEA2/6 (46 percentage points) ($p<.001$, LSD = 15.9). In the cool store there was no significant decrease in any of the treatments.

As more novel endophyte/grass combinations are released for commercial sale it is important to test each for compatibility and performance post-storage. The results of this study recommend controlled low-temperature, low-humidity storage to maintain endophyte viability.

Keywords: Endophyte, *Epichloë*, perennial ryegrass, *Lolium perenne*, storage

Acknowledgements

I would like to thank many people for their help and support during the time it has taken for me to complete this thesis.

- Pedro Evans, we 'put up with each other' for 8 years at DLF Seeds and Pedro offered honest opinion and guidance on not just the thesis, but, everything work related, and, sometimes not so work related. Even after I left DLF he was always just a phone call away when I had questions or just needed a sounding board. Sadly Pedro passed away near the completion of this thesis, it hasn't been easy without him.
- My wife, Deanna, for the 'gentle' nagging and support. Thanks for pretending to care about endophytes even though I know you really couldn't care less and 'just wanted me to finish the bloody thing'. It hasn't been an easy ride so thanks for sticking by me when the going got tough.
- My parents, Chris and Anthea, who supported me through the whole process and tried to take an interest in endophytes.
- My brothers, Tom and Harry, and extended family who wanted to ask me about what I was doing and even though I knew at times you regretted asking (the glazed over look gave it away) you still continued to ask. In particular Ainslie, Geoff, Lisa, Malcolm and Gen, Tim and Julie, Nana and Gran for always just asking how it was going.
- My supervisor, Michael McManus, who unfortunately passed away before I had finished. Thanks for taking an interest and providing useful ideas when I was just getting started.
- Cory Matthew, thank you for stepping in to get me back on track. It can't be easy coming in to supervise a student when they are already underway but you have done a great job and been enthusiastic about seeing me over the 'finish line'.
- Warren Williams, my co-supervisor, who has always been there in the shadows prodding me along and offering advice, even after all these years!
- My old workmates at DLF Seeds, Gavin and Garth, and my newer workmates at Plant and Food Research, Richard, Lester, Peter, Adam, Monika, Miriam and Linley, thanks for being sounding boards, proofreaders, stats helpers, asking me how things were going and, at times, the not so gentle encouragement to complete.
- DLF Seeds, thank you for the financial support and the resources to do the project. DLF Seeds gave me a great foundation to begin my career, as well as many life skills and I thank them for the opportunities given to me. In particular, Klaus Nielsen, who knew I could do it and did all he could from half a world away to keep me on track.

For Pedro

Table of Contents

List of tables

List of figures