Seed Factors Involved in Early Seedling Establishment of *Festuca arundinacea* (Tall Fescue)

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Abstract.

New Zealand pastures are commonly based on vigorously growing *Lolium* (ryegrass) species. In many situations, however, it is often advantageous to combine the vigorous qualities of *Lolium* species with the versatility of other species such as *Festuca arundinacea* (tall fescue) to result in a pasture which is high in quality all year. *F. arundinacea*, however, has poor seedling vigour which places it at a competitive disadvantage when sown with *Lolium*.

During seedling establishment, the seedling is dependent upon the food reserves present in the seed. The nature and amount of these reserves and the ability of the seed to mobilise them are therefore likely to have an affect on seedling performance. While much knowledge exists about the processes involved in seed reserve mobilisation in some cereals, little information is available for pasture grasses.

An in depth investigation of the behaviour of one seed lot of *F. arundinacea* was undertaken in order to gain a more detailed understanding of germination, reserve mobilisation and establishment processes in pasture grasses and how they relate to the processes of seedling growth. Comparisons between this species and *Lolium multiflorum* (Italian ryegrass) were undertaken throughout the study.

Germination and seedling growth of the *F. arundinacea* seed lot was found to be heterogenous and slower than in *L. multiflorum* due to later radicle emergence. Mobilisation of reserves and the onset of α-amylase activity correlated well with the utilisation of reserves in *L. multiflorum*. In both prechilled and non prechilled *F. arundinacea* seeds, however, anomalies were identified in the process which indicated that reserve mobilisation was less tightly coupled to seedling growth in *F. arundinacea*. Reciprocal plot analyses indicated that the beneficial effects of prechilling in most seeds of the *F. arundinacea* seed lot were not related to residual dormancy but were a thermal time benefit. It appears prechilling was allowing rate limiting steps in embryo growth to be advanced before visible germination. Apart from this, no real differences were detected in the way the two species mobilised reserves.

Differences in the appearance of α-amylase isoenzymes of *F. arundinacea* at different times during germination indicated that gene expression may be under some complex differential control mechanisms during germination and reserve mobilisation. Prechilling was not found to change the spectrum of isoenzymes, but merely to advance the time-course in which different isoenzymes appeared. There were also distinct differences in α-amylase isoenzyme patterns between *F. arundinacea* and *L. multiflorum*, and also wheat. Preliminary studies indicated that exogenous gibberellic acid was more effective in promoting α-amylase production in *L. multiflorum* than in *F. arundinacea*. However, α-amylase production in *F. arundinacea* was more susceptible to promotion by prechilling.

This study has identified a wide range of variables impacting on germination and seedling establishment in *F. arundinacea*. This, together with the lack of previous detailed studies on grass seed germination and seedling growth and the lack of literature on *F. arundinacea* germination in particular, highlights the enormity of the task ahead of extending key areas of this study to different seed lots and species.

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Abbreviations

ABA	- Abscisic Acid
cv.	- Cultivar
d	- Day
ER	- Endoplasmic Reticulum
GA_3	- Gibberellic Acid
h	- Hour
IEF	- Isoelectric Focusing
PGR	- Plant Growth Regulator
SE	- Standard Error
T_{50}	- Median Germination Time
T_{90}-T_{10}	- Uniformity of Germination

1. Introduction

1.1 Context of study

New Zealand pastures are dominated by species of vigorously growing *Lolium* which establish rapidly and are able to tolerate heavy grazing under a variety of environmental conditions. However, *Lolium* cannot tolerate extreme climatic conditions such as hot, dry summers or cold winters (Langer 1990). Therefore it is desirable to introduce some other grass species into *Lolium* pasture to complement its growth characteristics. One such option is *Festuca arundinacea* which is slow to establish but is an attractive pasture grass as it is hardy. It can grow on shallow, droughty soil and can also withstand waterlogging, flooding and considerable grazing (Barnes 1990).

In difficult climates it is often advantageous to combine the vigorous qualities of *Lolium multiflorum* (Italian ryegrass) with the versatility of *F. arundinacea* (tall fescue) to result in a pasture which is high in quality all year. However, the vigour of *L. multiflorum* which is advantageous in many situations can jeopardise the establishment of other species in mixed pastures (Langer 1990). *F. arundinacea* has poor seedling vigour which places it at a competitive disadvantage in mixed sowings (Charles 1965; Rhodes 1968). Therefore it is strongly recommended not to sow *F. arundinacea* with *L. multiflorum* (AgResearch 1993).

A study by Brock *et al.* (1982) has shown that after 10 days germination, *Lolium perenne* (perennial ryegrass) seedlings utilise 47% of their endosperm reserves compared with only 14% for *F. arundinacea* seedlings. The ideas that emerged from this study were that these differences in vigour were either due to relative growth rate differences, or that there were differences in the mobilisation process of the endosperm. However, as will be discussed in Chapter 2, this study had severe limitations and needs re-examination.

At these early stages of germination and seedling growth the seedling is dependent upon the food reserves present in the seed. The nature and amount of these reserves and the ability of the seed to mobilise them are therefore likely to have an affect on seedling performance. While much knowledge exists about the processes involved in seed reserve mobilisation in cereals, little information is available for pasture grasses. In addition, these reserve mobilisation processes are based on only a few species such as barley and wheat.

Reserve mobilisation of barley involves the synthesis and secretion of gibberellins by the scutellum. This stimulates a response by the aleurone layer to synthesise α-amylase and other hydrolytic enzymes *de novo* and to secrete them into the endosperm where they are involved in the mobilisation of starch reserves (Fincher 1989). Many differences have been observed between the model system of seed reserve mobilisation processes in barley and other cereal species. These include species variations in aleurone tissue responsiveness to added gibberellin, the presence of α-amylase inhibitor activity and the spectrum of α-amylase isoenzymes (Cornford and Coolbear 1992). It would therefore be dangerous to assume that these processes in pasture grasses would be identical to that described for barley. There is an obvious need for the range of activities and species studied to be broadened.

1.2 Objectives and experimental approach

This project aimed to determine whether reserve mobilisation in general and α-amylase activity in particular are limiting components of seedling growth and establishment in *F. arundinacea*. The objectives of the study were:

- to identify the optimum germination conditions of *F. arundinacea*, enabling differences between *L. multiflorum* (which has been the focus of previous research in this laboratory) and *F. arundinacea* to be identified.

- to compare changes in physical characteristics of *F. arundinacea* and *L. multiflorum* as germination and seedling growth proceed.

- to measure the activity of α-amylase in *F. arundinacea* and *L. multiflorum* during these stages.

- to characterise α-amylase isoenzymes present at different stages of germination in the two species.

-to try to identify mechanisms for the differences during germination and seedling growth in *F. arundinacea* and *L. multiflorum.*

Different cultivars and even different seed lots have been found to vary enormously in their germination characteristics and behaviour (Anslow 1964; Elgersma and Sniezko 1988; Flintham and Gale 1988). This is of concern in relation to selection of seed for testing procedures (Wiesner and Grabe 1972). This project could have been carried out in two ways - 1) a relatively superficial comparative study involving many seed lots and cultivars of each species, or 2) a more in depth approach with just one seed lot of each type. Due to interest in fundamental processes the latter approach was chosen. An obvious limitation is that 1) still needs to be done, however, the data from this project is intended to highlight key aspects that need to be addressed in a multi-seed lot survey.

1.3 Organisation of thesis

To meet the above objectives, this thesis has been organised into five chapters. Following and expanding on this introduction, Chapter 2, includes the description of relevant literature. The methods and materials of experiments performed to meet the above objectives are described in Chapter 3. Results and discussion of these experiments have been written up in three chapters, 4) Optimisation of *F. arundinacea* germination conditions, 5) Comparative time-course studies between *F. arundinacea* and *L. multiflorum*, and 6) Hormone responsiveness. The separate discussions of these results are tied together in a general discussion in Chapter 7.