

## **Insights into fine root development patterns of Perennial Ryegrass (*Lolium perenne* L)**

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### **ABSTRACT**

Since the initial report that roots finer than a given diameter are determinate in growth habit, little research has been done on the concept of diameter mediated growth patterns. An investigation into this phenomenon with a common pasture grass (*Lolium perenne* L.; Perennial ryegrass [Lp]), using very high resolution imaging (effective pixel size < 5 micron), provides some additional insights. Cultivars of this cross pollinating species have extensive variability in rooting patterns. Several different types of fine lateral roots (<0.5 mm diameter) have been documented: Long indeterminate, short indeterminate, long determinate, short determinate. Short vs. long determinate appears a plant determined characteristic since mixtures have not been observed. Only long indeterminate lateral roots have lateral branching. There appear to be two types of long indeterminate roots, frequent and atypical. The frequent type of indeterminate lateral root are typically half the diameter of the parent root. The atypical type may be as large as the parent root diameter, and may appear at any location along the parent root. Short indeterminate roots differ from long ones in not having branching, growing slowly, and being smaller in diameter.

**KEYWORDS:** Fine Roots, Developmental Patterns, Indeterminate, Determinate, Short Laterals

### **INTRODUCTION**

Perennial Ryegrass (PR - *Lolium perenne* L.) is a common species in both turfgrass and pastures. Although true breeding lines exist, most PR "cultivars" start as mixtures of lines and are increased through bulk population seed increase. The end result is a genetic mixture that may have been selected for some shoot characteristics, but not directly selected for root characteristics. To directly select for rooting characteristics, knowledge of the morphological characteristics of the subject root system and an understanding of its growth pattern are needed. To our knowledge, these databases do not exist, nor is there documentation of variability in rooting patterns amongst seedlings in a given cultivar of perennial ryegrass..

### **MATERIALS AND METHODS**

Seeds of perennial ryegrass cv BG34 (*Lolium perenne* L.), were germinated on paper towels and transplanted into aeroponic scanning meso-rhizotrons (ASM – see description below) at 5 seedlings per ASM. Images of the root systems were captured daily for 60 days. Apparent differences in rooting patterns (lateral branching density, length, growth duration, and, finally, diameter) were noted. Twelve seedlings were selected for cloning, and, after two clonal divisions, clonal plants were separated into individual tillers and planted into ASMs at six tillers per ASM, 12 clones, eight replications, repeated three times. Images were acquired daily for 54 days. After the second repeat, five clones with distinct rooting patterns (Table 1), were selected for a soil experiment.

### **Experimental design**

Tillers from the five clones were planted in 10 cm pots, using un-sterilized, limed, gilpin silt loam soil (fine-loamy, mixed, mesic Typic Hapludults, pH 7.10), with four replications, and repeated three. At the end of a 60 day growth period, WinRhizo was used to determine root length and diameter.

### **Microscopic analysis**

Microscopic analysis (Zeiss Discovery V.12 Stereo at 100x with a Lovins Field Finder DX00877 measurement slide), of roots from the third repeat of the experiment, was used to acquire exact diameter measurements.

### **Aeroponic scanning mesorhizotrons.**

To construct ASMs, Epson Perfection V350 scanners (Epson.com) were modified by sealing (water proof) the glass platten, surrounding it with 1 cm thick closed pore foam, and adding a light proof lid. The upper long side of the foam surround was slit to hold individual plants. Inlet and outlet tubes of 1 cm diameter PVC tubing were inserted along the short edges, and nutrient fog (0.125 x Hoagland solution (Hoagland and Arnon, 1954) aspirated with a Humidistat (Humidistat Corp. Boca Raton FL) fogger) blown through the resulting chamber. The scanners were placed in holders with the glass platten at 60 degrees from horizontal so that the roots could grow along the platten surface. The scanners were automatically activated by a computer program which then captured the resulting images.

## **RESULTS**

Five distinct types of lateral root were found: short determinate, long determinate, short indeterminate, long indeterminate, and atypical indeterminate. A given clone had only one of these types of lateral, except for the atypical laterals which appeared infrequently on all clones. Only long and atypical indeterminate lateral roots developed secondary laterals. There were three distinct diameter classes of root: shoot borne – 0.436 +/- 0.025 mm; lateral – 0.123 +/- 0.005 mm; and secondary lateral – 0.082 +/- 0.005 mm. Mean diameter of these classes differed significantly between clones. The scanning resolution used for the WinRhizo data, 94  $\mu\text{m}^{-1}$  was too low to separate the secondary lateral peak from the lateral peak; a resolution of 189  $\mu\text{m}^{-1}$  will be needed.

Modeling root diameter class distributions of the whole root system was best with an "extreme value" non-linear regression model (EVL), while a double Gaussian non-linear regression (DGL) model, gave the best fit to individual shoot borne root data. The best fit for predicting root development pattern is to model individual shoot borne roots and their laterals with a DGL model and then pool these to get whole root system data which will then be an EVL best fit.

## **DISCUSSION**

A database with lateral root type, lateral root density on the shoot borne root, and average diameter class for shoot borne, lateral and secondary lateral roots for different clones of perennial ryegrass has been developed for five clones of cv BG34. To model root system development from these datasets a double Gaussian non-linear is appropriate. If resolutions of  $> 189 \mu\text{m}^{-1}$  are used then a triple Gaussian model should be appropriate.