THE EFFECT OF NITROGEN FERTILIZATION ON SHORT-TERM FINE ROOT DYNAMICS IN A BARRIER ISLAND DUNE COMMUNITY

by

John Walter Hutton
B.S.A. 1996, The University of Georgia, Athens

A Thesis Submitted to the Faculty of Old Dominion
University in Partial Fulfillment of the Requirement for
the Degree of

MASTER OF SCIENCE

BIOLOGY

OLD DOMINION UNIVERSITY
May 2001

Approved by:

Frank P. Day (Director)

Kneeland Nesius (Member)

Gerald Levy (Member)

ABSTRACT

THE EFFECT OF NITROGEN FERTILIZATION ON SHORT-TERM FINE ROOT DYNAMICS IN A BARRIER ISLAND DUNE COMMUNITY.

John Walter Hutton
Old Dominion University, 2001
Director: Dr. Frank P. Day

Fine root dynamics are an important yet poorly understood component of terrestrial ecosystems. In recent years a number of researchers have focused on fine root dynamics; their work has looked at patterns across one or multiple growing seasons on a scale of monthly measures. The purpose of this study was to add to the overall understanding by looking at fine root dynamics on a shorter temporal scale. Minirhizotron observation tubes were used to assess the effect of nitrogen fertilization on short-term fine root dynamics in a nutrient limited, barrier island dune system. Root length elongation and mortality rates followed expected patterns with soil depth, with the highest values for both occurring in the upper 22 cm. Unexpectedly, soil nutrient analyses did not confirm decreasing nitrogen levels with increasing depth although high variability suggests an inadequate sample size. Total % nitrogen was significantly increased by the fertilization regime and corresponded to significantly higher root elongation and mortality rates. However, mortality rates (0.61 mm cm^{-2} day^{-1}) greatly exceeded root length elongation rates (0.18 mm cm^{-2} day^{-1}) in both control and treatment plots. These results support one of the two hypotheses summarized by Hendricks et al. (1993a), namely that fine root turnover rates increase with nitrogen availability. The effect of N fertilization was much more pronounced on mortality rates than on root length

elongation rates. This is suggestive of a belowground die back in response to nitrogen fertilization, perhaps associated with a shift in allocation to aboveground production.

ACKNOWLEDGMENTS

I would like to thank Dr. Frank P. Day for his guidance and support throughout the project. I also appreciate the input and guidance of the other members of my committee, Dr. Gerald Levy and Dr. Kneeland Nesius. Thanks go to Jimmy Spitler and Randy Carlson for driving me out to the island and helping me gather GPS data. For forsaking his own work to help and feed all the other members of the lab, I would especially like to thank John Dilustro. Additionally, I am in debt to LeRoy Rodgers and Ed Crawford for their help in the field as well as in the laboratory. Lastly, I would like to thank my wife, Erika, for all her support throughout my Master's program. This work was supported by University of Virginia subcontract 5-26173 on the VCR-LTER grant from the National Science Foundation.

TABLE OF CONTENTS

Page

LIST OF TABLES

LIST OF FIGURES

[?]INTRODUCTION

Barrier islands have long been known for their distinctive vegetation. The grasses of the sand dunes, the wind-pruned appearance of the trees and the distinct zonation patterns of these systems have been the subject of much ecological research (Ehrenfeld 1990). These ecosystems are limited by a number of extreme environmental factors, such as salt spray, salt water, moving sand and poor soil (Ehrenfeld 1990). Salt spray and salt water can be sources of stress and mortality to barrier island vegetation (Wells and Shunk 1938). However, spray may also constitute an input of nutrients, other than nitrogen and phosphorous, to these nutrient-limited systems (Van der Valk 1974).

Other studies have confirmed nitrogen limitation in Virginia barrier island soils (Conn and Day 1992, Day 1996) as well as other coastal dune ecosystems (Kachi and Hirose 1983). It has been suggested that this deficiency is a limiting factor for growth in most dune plant species (Kachi and Hirose 1983). Various studies have found that the addition of nitrogen fertilizer to dune soils leads to increased total plant biomass (Day 1996, Kachi and Hirose 1983, Willis 1965).

As dune soils are made up primarily of sand, they generally have very low cation exchange capacities and therefore poor nutrient retention. These soils are almost always very low in organic matter, nitrogen, phosphorous and potassium (Ehrenfeld 1990). The low nitrogen status of barrier island sandy soils makes nitrogen availability easy to manipulate. Also the sandy soils of the islands make root observation relatively easy, providing for an excellent environment in which to observe nutrient limited root

---

[?] Journal model is *Plant and Soil*.

dynamics (Weber and Day 1996).

Studies in varied ecosystems have suggested that greater than one-half of net primary productivity is allocated belowground (Eissenstat and Yanni 1997, Fogel 1985, Grier et al. 1981). In addition, carbon expended for root maintenance often exceeds that used for root production (Eissenstat and Yanni 1997). Also, nutrient inputs to the soil from root systems can be equal to or greater than that of aboveground litterfall (Arthur and Fahey 1992, Vogt et al. 1986). Thus, root production and turnover have direct consequences for carbon and nutrient cycling. However, root system dynamics are a poorly understood aspect of terrestrial ecology due to the technical difficulties inherent in their study.

Several researchers, in varied ecosystems, have observed either no change or a decrease in absolute root biomass as a result of nitrogen fertilization (Wilson and Tilman 1991, Birk and Vitousek 1986, Bowman et al. 1993). However, plants in nutrient poor environments tend to allocate proportionally more carbon to roots than shoots (Chapin 1980, Gleeson and Tillman 1990, Vitousek and Sanford 1986). It follows that a release from nutrient constraints would often lead to a decrease in root carbon allocation in proportion to shoot allocation. While there may be a shift in proportional allocation, root production might still be stimulated by nutrient augmentation as many studies have shown (Gleeson and Tillman 1990, Stevenson and Day 1996, Tilman and Wedin 1991).

Hendricks et al. (1993) summarized two contrasting hypotheses concerning the effect of N availability on fine root carbon allocation. The first suggests that relative fine root carbon allocation decreases and that longevity and turnover rates are not significantly affected by increasing N availability. The second hypothesis maintains that relative fine root carbon allocation remains constant and that fine root turnover rates increase with nitrogen availability.

Weber and Day (1996) found that nitrogen fertilization resulted in increased root length density in a barrier island dune ecosystem. Pregitzer et al. (1993) found that root longevity increased under high nutrient conditions in a forested ecosystem. To date, little work has been done to determine the effect of nitrogen fertilization on short-term root length elongation and mortality rates.

Research on plant root systems in situ is technically difficult due to the fact that soil limits their accessibility for observation (McMichael and Taylor 1987). A number of techniques have been developed to study root production and turnover: radio-tracer techniques (Dalhman 1968), the carbon budget method (Raich and Nadelhoffer 1989), the nitrogen budget method (Nadelhoffer et al. 1985), the ingrowth core method (Neill 1992), sequential coring (Powell and Day 1991), the rhizotron chamber method (Bohm 1979), and the minirhizotron method (Hendrick and Pregitzer 1992). Of these, only the rhizotron and minirhizotron techniques offer non-destructive means of examining root dynamics. The rhizotron method, however, has several distinct disadvantages. First, since the conditions in the rhizotron are not completely natural, the results from experiments using this technique may differ substantially from those of field conditions (Cheng et al. 1991). Second, construction costs for rhizotron facilities can be quite high (Cheng et al. 1991).

Minirhizotrons are clear butyrate tubes that have been used in a number of ecosystems to videotape roots, in situ, for later analysis (Hendrick and Pregitzer 1992, 1993b, 1996, Upchurch and Ritchie 1983, Weber and Day 1996). However, one serious limitation to this technology is the inability to measure biomass directly. Several researchers are currently developing allometric equations to allow estimation of biomass with minirhizotrons (John Dilustro *personal communication*). However, these relationships will most likely be system dependent. Soil coring must be done in conjunction with minirhizotron work to examine biomass along with other root dynamics.

The current study quantified the root length elongation and mortality rates of a barrier island dune community. The work was actually a subset of a long-term study that has examined root dynamics on multiple temporal scales. The original study began in 1992 with an experiment examining fine root dynamics across a growing season using monthly tapings. Tubes have since been taped on a twice-yearly basis. The current study adds knowledge of fine-root dynamics on a smaller temporal scale. The primary objective was to determine how short-term root dynamics are affected by nitrogen fertilization on a nutrient poor, barrier island dune ecosystem using minirhizotron observation tubes.

MATERIALS AND METHODS

Site Description

Hog Island (37^{o}40' N, 75^{o}40' W) is a barrier island off the eastern coast of the Delmarva Peninsula on the Virginia Coast Reserve (VCR) Long-Term Ecological Research (LTER) Site in Virginia, USA (Fig. 1). This series of barrier islands is owned and managed by The Nature Conservancy. Although a small community existed on the southern end, Hog Island has been largely uninhabited since the late 1940's (Dueser et al. 1976). All cattle were removed from the island in 1980 (Hayden et al. 1991).

Accretion on the north end of Hog Island has produced distinct dune complexes as well as a foredune area while the south end has been eroding (Hayden et al. 1991). During 1995 and 1996, the north end of the island eroded but has since begun to widen again (John Porter, *personal communication*).

From the Atlantic Ocean to the bayside of the island, a chronosequence of dunes has been aged, with formation dates ranging from 1871 to 1985 (Hayden et al. 1991). The present study was located on the well-drained, 1955 dune line that is bordered to the east by a freshwater marsh of *Spartina patens* Muhl. and *Distichlis spicata* (L.) Greene and to the west by thickets of *Myrica cerifera* L. (Fig. *2*). The dominant plant communities on this ridge are the perennial grasses, *Ammophila breviligulata* Fern., *Spartina patens* Muhl. and *Panicum amarum* Ell. (Table 1). The soil is a Newhan series within a Newhan-Corrolan complex that provides few nutrients and exhibits low nutrient and water retention (Duesser et al. 1976).

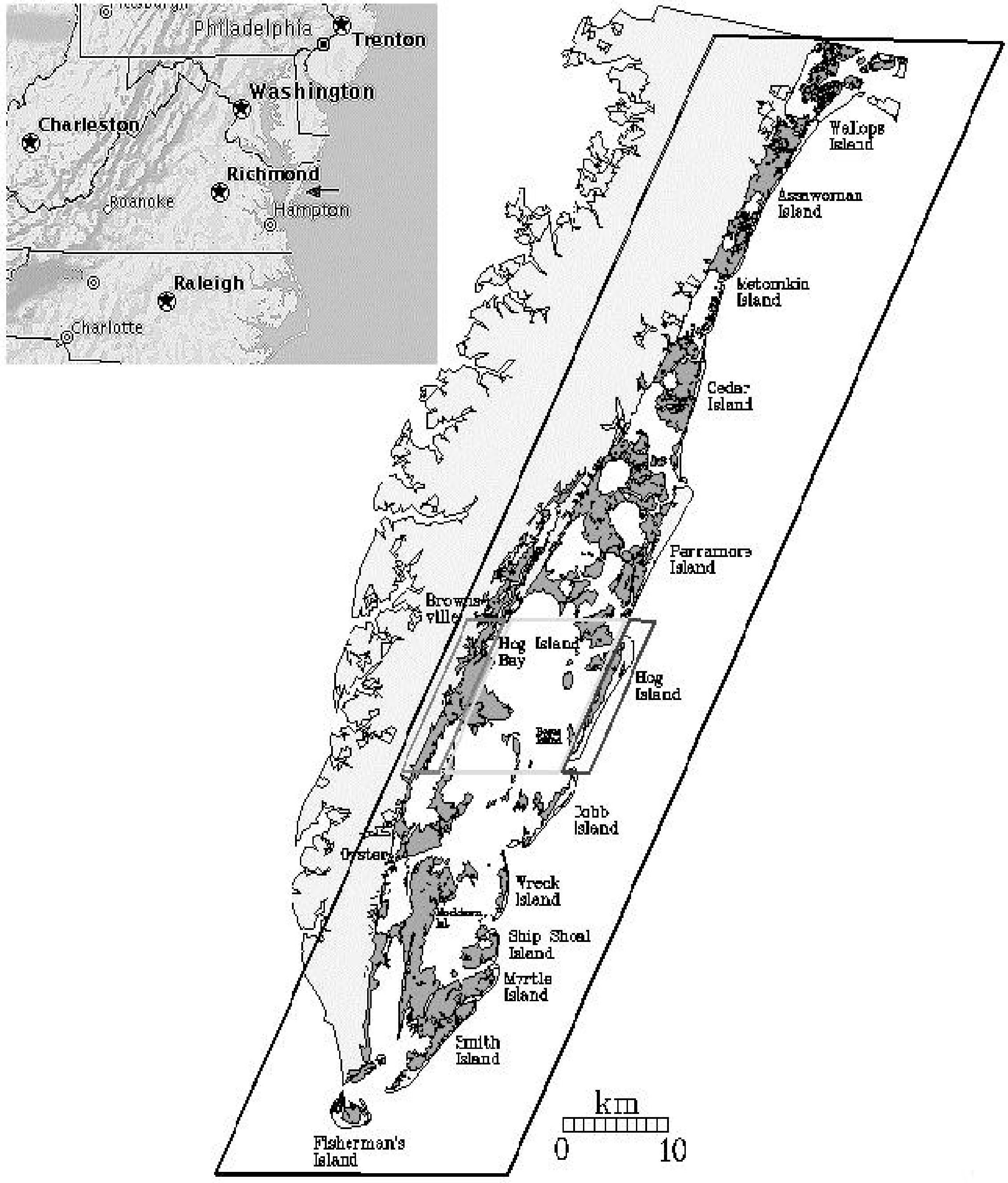


Fig. 1. Map of Virginia Coast Reserve; research site is on Hog Island

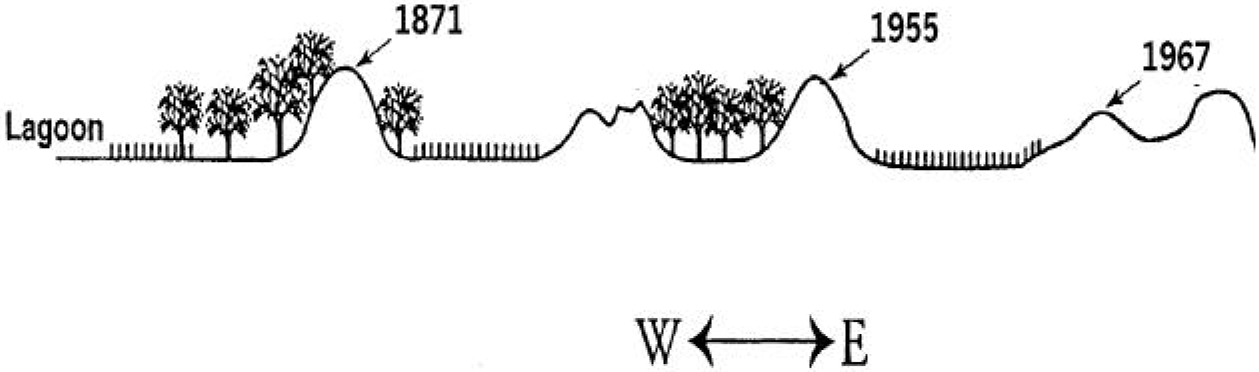


Fig. 2. Diagam of the dune chronosequence on Hog Island; study plots are on the 1955 dune li

Table 1. Plant species in both fertilized and control plots ranked by mean % cover (data from two quarter meter square quadrats in each of 4 plots sampled per treatment 5/99). Ambr = *Ammophila breviligulata* Fernald, Paam = *Panicum amarum* Ell., Sppa = *Spartina patens* (Aiton) Muhl., Soca = *Solanum carolinense* L., Scsc = *Schizachyrium scoparium* (Mich.) Nash, Ruac = *Rumex acetosella* L., Casp = *Carduus spinosissimus* Walter.

Fertilized Plots		Unfertilized Plots	
Species	% Cover	Species	% Cover
Ambr	83.75	Sppa	34.17
Sppa	45.0	Scsc	14.17
Ruac	16.88	Paam	12.5
Paam	13.13	Ambr	7.5
Scsc	0	Ruac	5.0
Casp	0	Casp	2.5
Soca	0	Soca	0.83

Sampling Design

Eight 3 X 3 meter plots, spaced a minimum of six meters apart, were selected as representative of the dune ecosystem (Weber and Day 1996). They were then randomly divided into four control and four experimental plots (Weber and Day 1996). One of the control plots has since been removed from the study due to the effects of encroachment by *Myrica cerifera* (Weber and Day 1996). Fertilized plots received three applications, during the first year of the study, of 15 g N m^{-2} in the form of a 70%-30% mix of coated temperature-release urea to uncoated urea (Weber and Day 1996). Since that time, they have received 15 g N m^{-2} once per year in the middle of the growing season. Four minirhizotron tubes were placed in each plot, perpendicular to and 1 meter from one of the adjacent sides (Fig. 3). In an effort to reduce edge effect, tubes were oriented with the etched frames facing the center of the plots (Weber and Day 1996).

Minirhizotrons

The minirhizotron tubes are 2 m long, clear butyrate tubes with an inner diameter of 5.08 cm and 0.65 cm thick walls (Weber and Day 1996). They have approximately 80 etched frames along the side to allow taping at the same location within the tube on all sampling dates (Weber and Day 1996). All tubes were painted with flat black paint to just below the soil surface, wrapped with black electrical tape, and capped with a PVC top to prevent rain and light entry (Weber and Day 1996). Once prepared, they were installed in the soil at an angle of 45^{o}. All tubes were installed between February 15 and 29, 1992 (Weber and Day 1996). They have since been used for monthly data collection

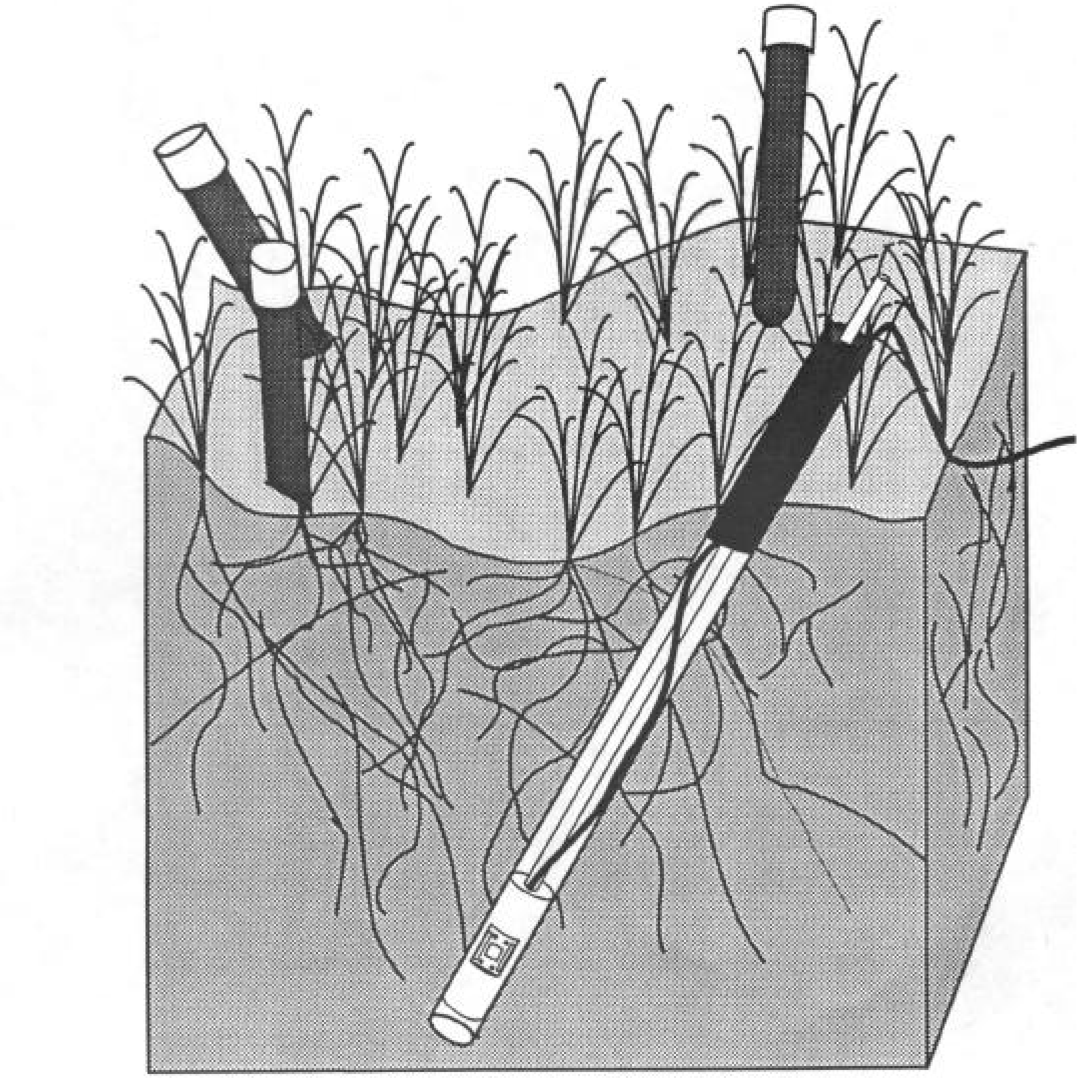


Fig. 3. Installation of tubes within plots. A minirhizotron camera is inserted into a tube.

in 1992 and twice-yearly observations over five years for a long-term study on the effects of nitrogen fertilization on root phenology.

Each minirhizotron was measured once every 6 days from May 21, 1998 to June 14, 1998, resulting in a total of 4 sampling intervals. The minirhizotron images were collected on a hi-8 videotape recorder with a Bartz Technology microvideo camera.

Image Analysis

A subsample of the total frames was selected in a stratified random manner with an equal number of randomly selected frames within each of three depth classes. During the first digitizing session, certain roots were selected non-randomly based upon their suitability for measuring root growth and mortality. Roots were deemed suitable if the tip of a root could be clearly seen within the etched frame. The individual roots within these frames were then followed through the course of the study.

Root images were analyzed using ROOTS software (ver 1.05, Michigan State University Remote Sensing Laboratory). Root length and average width were traced using a mouse and the measurements, along with the root coordinates and a numerical identifier, were then written to a database file by ROOTS. Roots were also classified by color as black, white, or dark although this information was not used in the analysis of the data.

Roots could then be identified and re-measured by using ROOTS to recall and overlay the tracings and identifiers from an image at time t-1 when analyzing images at time t (Hendrick and Pregitzer 1993b). After digitizing was completed, the files created by ROOTS were compiled into a single folder that was manipulated using Microsoft Foxpro (a database program).

Root Length Elongation

Root length elongation rates were calculated as the length at time t+1 minus the length at time t when the length at time t+1 was greater than zero. This included the change in length of all roots present at both time t and time t+1 as well as the length of new roots, or roots present at time t+1 but not at time t.

Mortality

Mortality rates were calculated as the length at time t+1 minus the length at time t when the length at time t+1 was equal to zero. This included the change in length of all roots that were present at time t but not at time t+1.

Data Analysis

A log transformation was used to normalize the root length elongation and mortality rate data. Tubes were nested in plots as in Hendrick and Pregitzer (1992). Frames were pooled into three roughly 20 cm vertical depth classes (0-22 cm, 22-46 cm, 46-68 cm). As depth class one was necessarily related to depth class two and depth class two to depth class three, depth was analyzed as a repeated measure. Dates were also analyzed as a repeated measure for this study. Thus, the ANOVA model had plots nested within treatments and depth class and time as repeated measures. Due to the imbalance of the design (control plot 2 was removed), the data had to be analyzed using a general linear models procedure as opposed to a nested procedure (SAS Institute 1985).

Soil Nutrient Analysis

Three 60 cm cores with 4 cm diameters were collected in each of the eight plots using a soil corer. The full core was separated into three depth classes (0-20 cm, 20-40 cm, 40-60 cm) with each depth class sample being placed into a separate labeled ziplock bag. These depth increments were chosen to correspond to the depth classes used in the analysis of the minirhizotron data.

The bagged soil samples were then transported to the laboratory and refrigerated for two days prior to sample preparation. Subsamples were taken from each sample using the cone and quarter method (Joseph Rule, *personal communication*) in order to best represent the whole sample. This method involved mixing each sample thoroughly within the bag, pouring the sample onto a clean surface in the shape of an inverted cone then taking one quarter of this cone as a representative subsample. Subsamples were placed in labeled manilla envelopes and oven dried for 36 hours at 50^0C. Time and temperature followed the methodology in Verardo et al. (1990) for analyzing sandy sediment samples for total N and C.

Dry samples were removed from the oven and ground to a fine homogenous consistency using a mortar and pestle. The ground samples were placed in clean, labeled envelopes and set in a desicator.

Each subsample was analyzed for percent total N using a Carlo Erba CHNS analyzer. Sample analysis procedures followed the methodology in Cutter and Radford-

Knoery (1991). Due to the intrinsically low N content of depauperate dune sandy soils, it was necessary to use the largest sample mass possible while still achieving a complete burn. After experimenting with several different sizes, I opted for a sample size of approximately 15 mg, which corresponds closely to the mass used by Verardo et al. (1990). After the 15 mg sample was weighed in a tin capsule, approximately 10 mg of vanadium pentoxide was added to the top of the sample. The capsule was then folded into a ball small enough to drop through the aperture into the combustion column of the CHNS analyzer.

RESULTS

Root Length Elongation

The results showed the three main effects to be significant (Table 2). Values ranged from 0.47 mm cm^{-2} day^{-1} for the first time interval in the fertilized treatment to 0.02 mm cm^{-2} day^{-1} for the final time interval in the control treatment (Fig. 4). Variation was also much greater for the fertilized treatment group. The main effect of depth was shown to be significant (F=17.05, p=0.0001). Similar depth patterns of root length elongation can be seen in both treatments with the highest values occurring in the 0-22 cm depth range.

Fig. 5 shows the pronounced effect of the fertilization treatment on root length elongation (F=8.34, p=0.0343). Values averaged across depths by treatment ranged from 0.35 mm cm^{-2} day^{-1} in the fertilized treatment to a low of 0.05 mm cm^{-2} day^{-1} in the control group. While the main effect of date was also significant (F=4.74, p=0.0052), only the first and last dates differed significantly (Tukey's, $P < 0.05$).

Mortality

The results showed two main effects and one interaction to be significant (Table 3). Mortality rates for all depths and dates for both the fertilized and control treatments ranged from 1.8 mm cm^{-2} day^{-1} for the first time interval in the fertilized treatment to 0.09 mm cm^{-2} day^{-1} for the third time interval in the control treatment (Fig. 6). Depth was shown to have a significant effect on mortality rates (F=72.72, p=0.0001), with the highest values occurring in the 0-22 cm group. The depth*treatment interaction was

Table 2. Nested doubly repeated measures analysis of variance examining the effect of fertilization on root length elongation over time across three depth classes. DF = Degrees of freedom, SS = Sum of squares, F Value = calculated F value, Adj Pr > F = Greenhouse-Geisser adjusted F value.

Source of variation	DF	SS	Type III MS	F value	Pr>F
Treatment	5	0.212	0.0424	8.336	0.0343
Error	5	0.0255	0.0051		
Plot(Treatment)	5	0.0255	0.0051	2.464	0.0438
Depth class	2	0.0352	0.0021	17.05	0.0001
Date	3	0.0294	0.0098	4.741	0.0052
Depth class*Date	6	0.0045	0.00075	0.363	0.8991
Treatment*Depth class	2	0.0078	0.0039	1.91	0.1578
Treatment*Date	3	0.0096	0.0032	1.547	0.2128
Treatment*Date*Depth class	6	0.0027	0.00045	0.217	0.9697
Error	83	0.3122	0.0021		

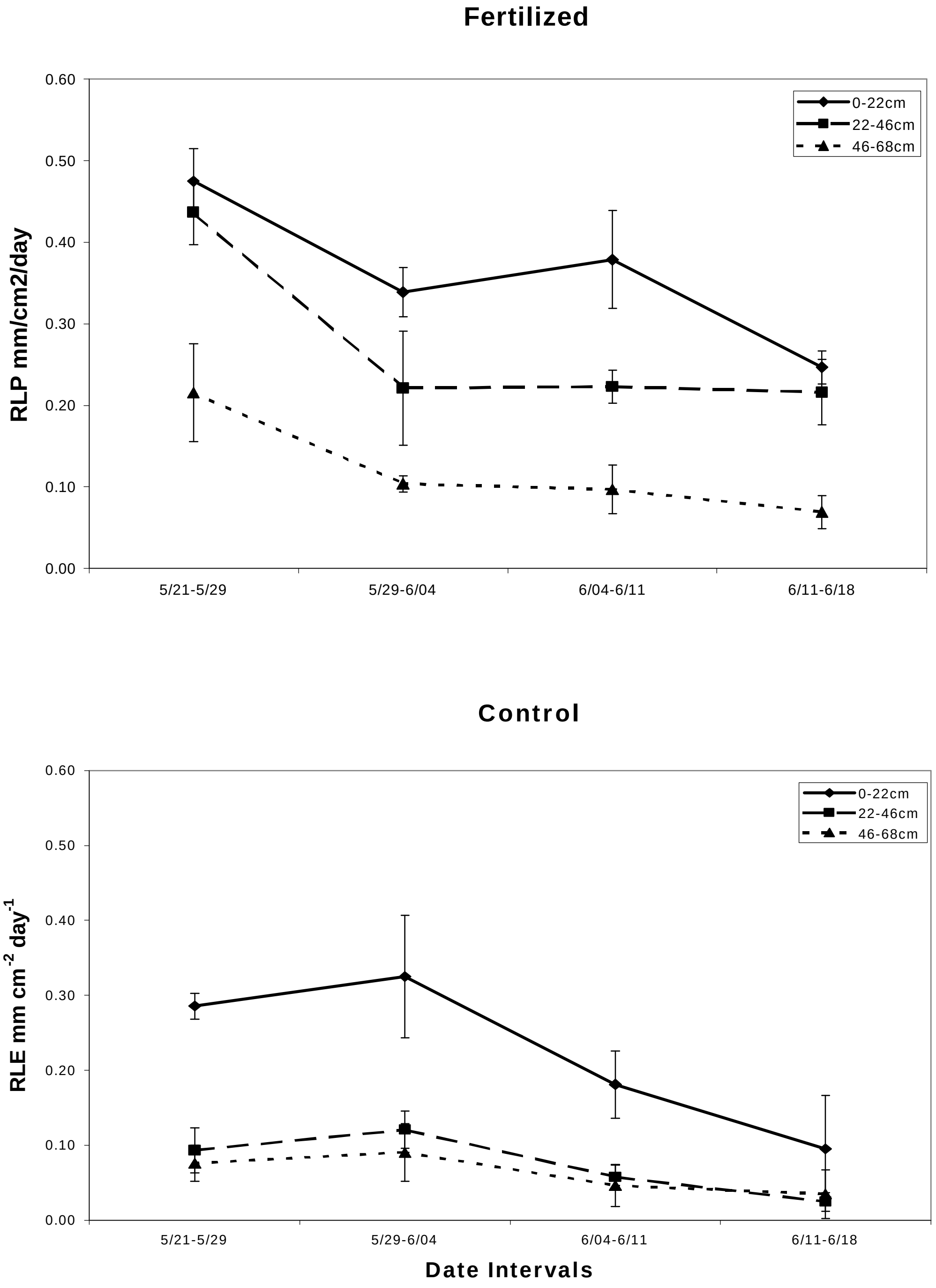


Fig. 4. Root length elongation (RLE) for fertilized and control treatments for all three depth classes from 5/21/98 – 6/19/98 (Mean and standard error of the mean).

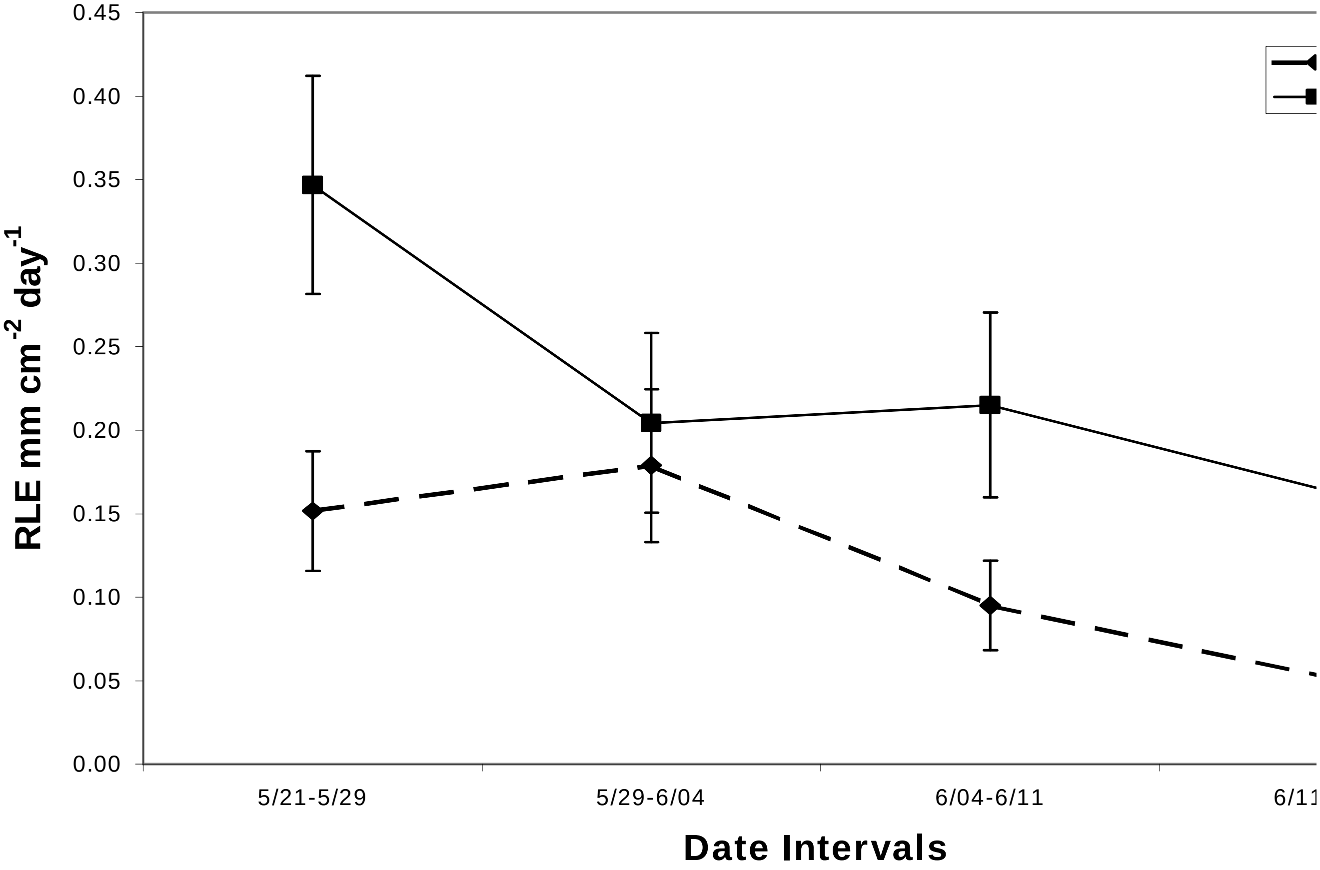
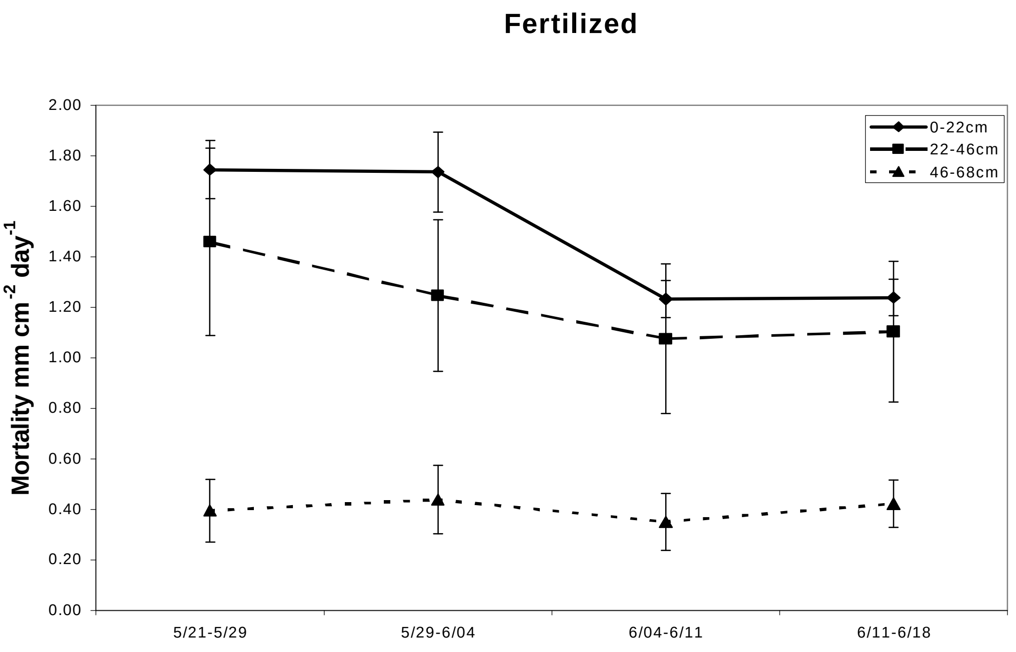


Fig. 5. Root length elongation (RLE) (change in length of roots from time t to time t+1) for fertilized treatments with all depths pooled from 5/21/98 – 6/19/98 (Mean and standard error of the mean).

Table 3. Nested doubly repeated measures analysis of variance examining the effect of fertilization on root mortality rates over time across three depth classes. DF = Degrees of freedom, SS = Sum of squares, F Value = calculated F value, Adj Pr > F = Greenhouse-Geisser adjusted F value.

Source of variation	DF	SS	Type III MS	F value	Pr>F
Treatment	1	8.4360	8.4360	36.7600	0.0018
Error	5	1.1480	0.2295		
Plot(Treatment)	5	1.1480	0.2295	5.6690	0.0003
Depth class	2	5.8880	2.9441	72.7200	0.0001
Date	3	0.1767	0.0589	1.4540	0.2372
Depth class*Date	6	0.0045	0.0229	0.5666	0.7551
Treatment*Depth class	2	0.6130	0.3065	7.5710	0.0012
Treatment*Date	3	0.1074	0.0358	0.8832	0.4555
Treatment*Date*Depth class	6	0.0396	0.0066	0.1624	0.9856
Error	55	2.2270	0.0405		



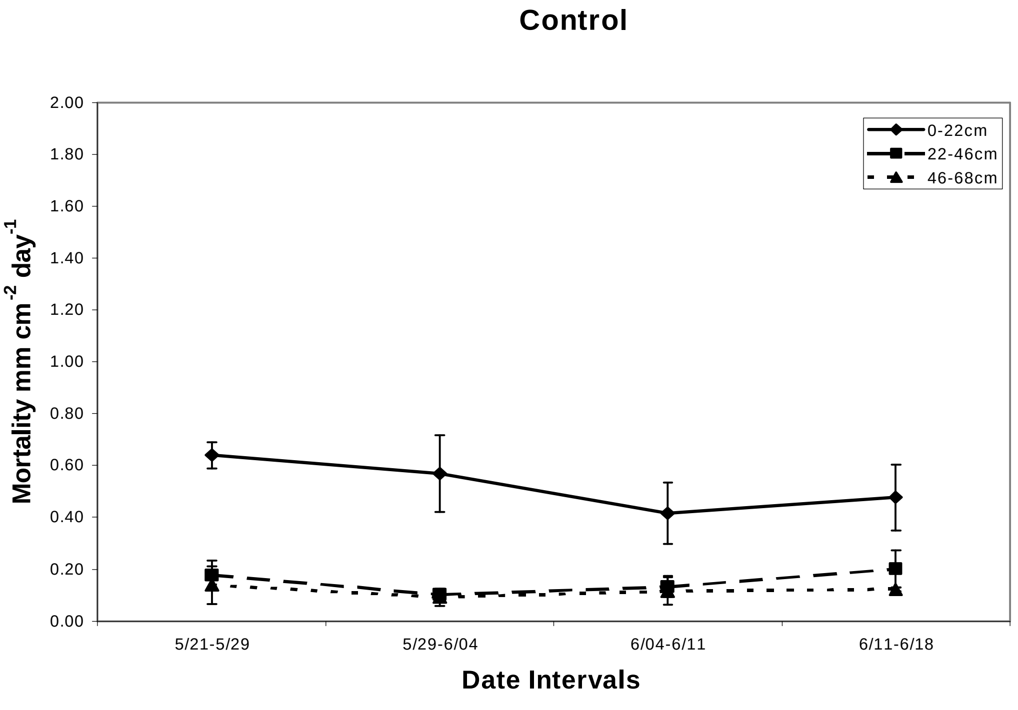


Fig. 6. Mortality rates for fertilized and control treatments for all three depth classes from 5/21/98 – 6/19/98 (Mean and standard error of the mean).

significant (F=7.57, p=0.0012). The 22-46 cm depth class seems to have shown a greater response to the fertilization treatment than the other two depth classes (Fig. 6).

Fig. 7 shows the pronounced effect of the fertilization treatment on mortality rates (F=36.76, p=0.0018). Values averaged across depths by treatment ranged from 1.1 mm cm^{-2} day^{-1} in the fertilized treatment to a low of 0.25 mm cm^{-2} day^{-1} in the control group.

Root Length Elongation vs. Mortality

Mortality rates were greater than root length elongation rates for both the control and fertilized treatments. Mortality rates, averaged across depths, ranged from 1.10 mm cm^{-2} day^{-1} in the fertilized treatment to a low of 0.22 mm cm^{-2} day^{-1} in the control plots (Fig. 8). Root length elongation values, also averaged across depths, ranged from a high of only 0.38 mm cm^{-2} day^{-1} in the fertilized treatment to a low of 0.05 mm cm^{-2} day^{-1} in the control plots (Fig. 8). Fig. 8 shows the much greater response of mortality rates to the fertilization treatment than that of root length elongation. Mortality rates for the fertilized plots were approximately triple that of the control plots, whereas root length elongation rates less than doubled.

Soil Nitrogen Analysis

The results showed one main effect to be significant (Table 4). Only the main effect of treatment was significant (F=8.17, p=0.0061). The fertilized group had significantly higher nitrogen concentrations (mean=0.0145) than the control (mean=0.008). Variability was high for both the control and fertilized plots (Fig. 9), possibly masking significant trends by depth.

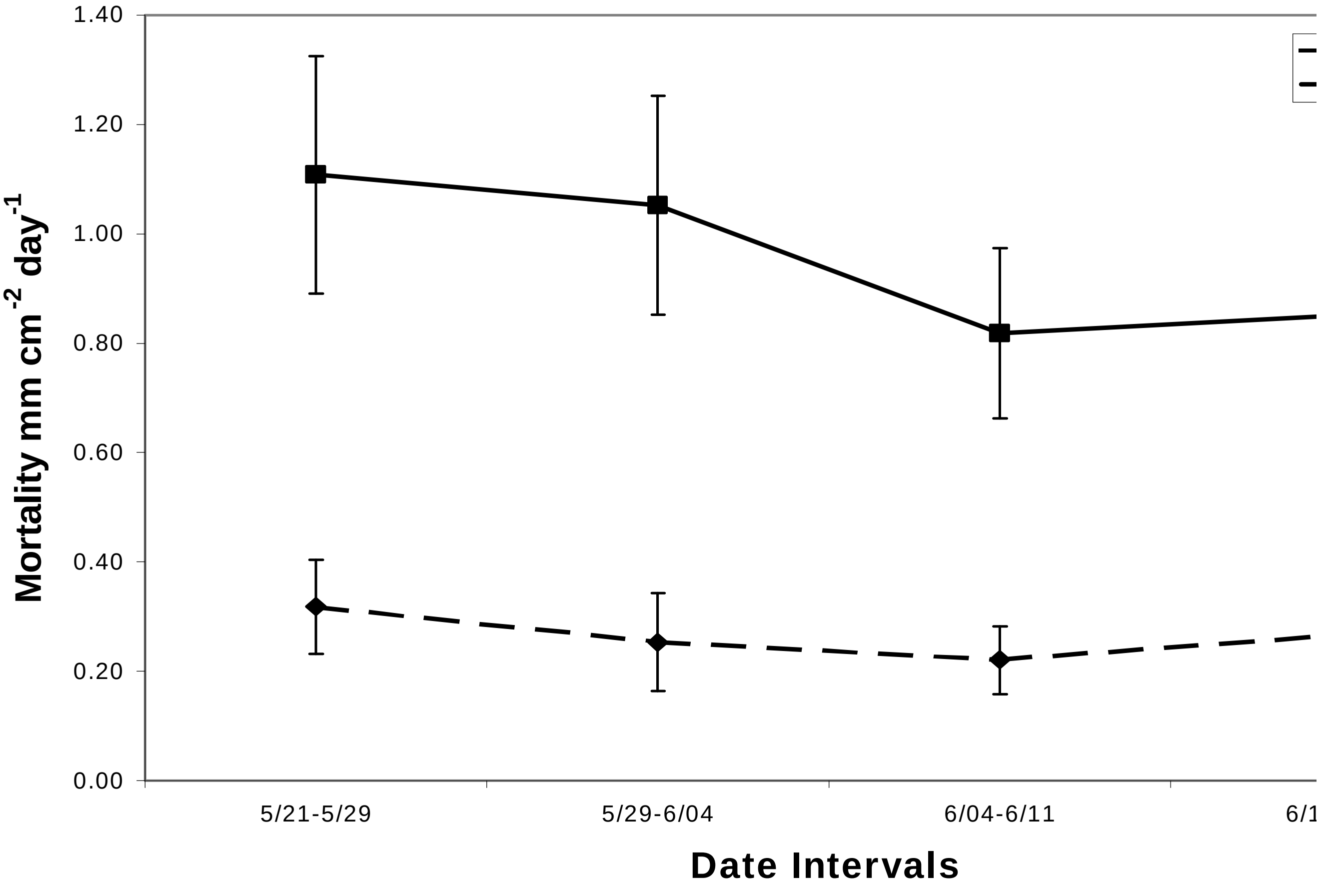


Fig. 7. Mortality rates for fertilized and control treatments with all depths pooled from 5/21/98 – 6/1
and standard error of the mean).

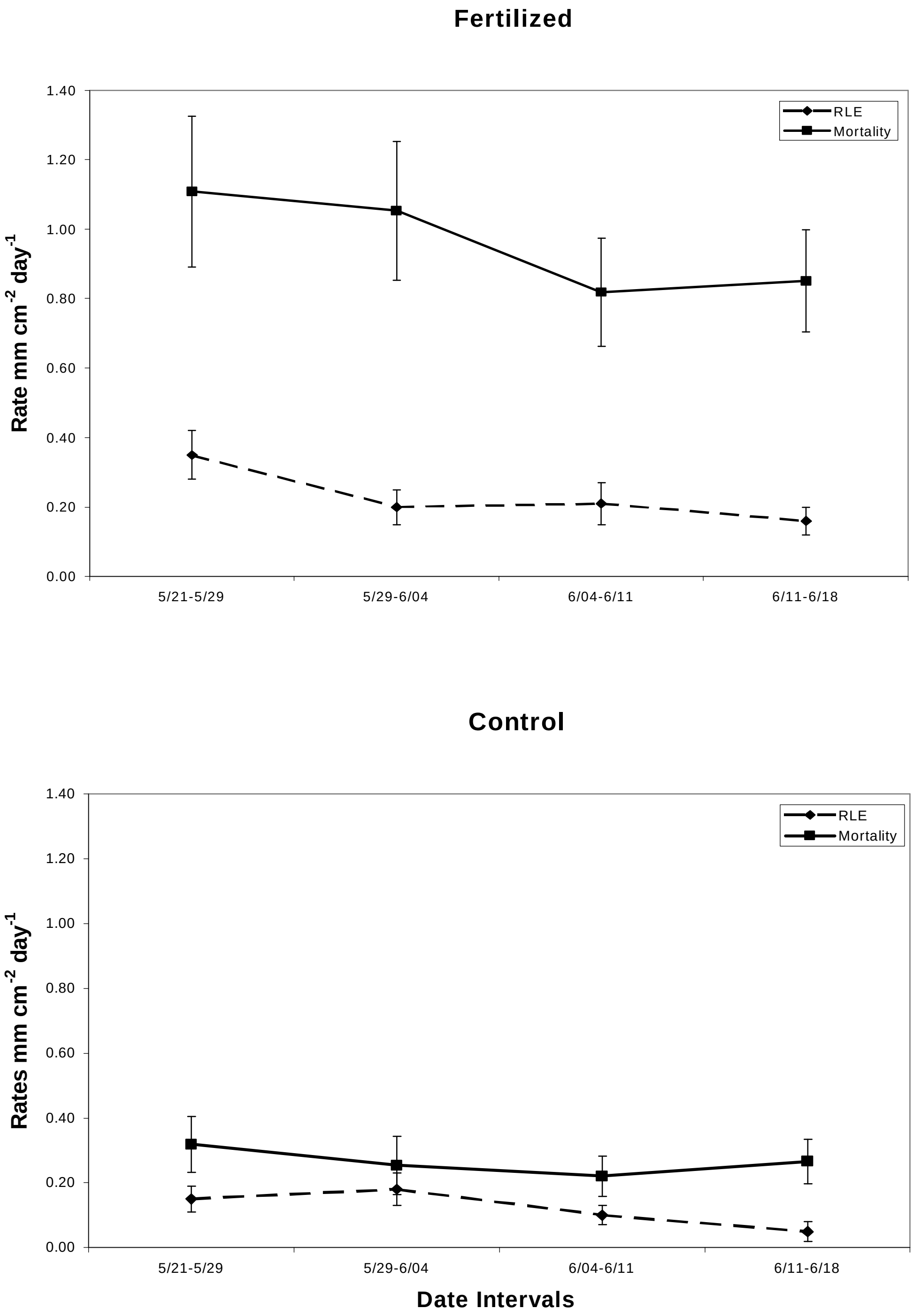


Fig. 8. Root length elongation vs. mortality rates for fertilized and control treatments with all depths pooled from 5/21/98 – 6/19/98 (Mean and standard error of the mean).

Table 4. Nested analysis of variance examining the effect of fertilization on % total nitrogen by mass across three depth classes. DF = Degrees of freedom, SS = Sum of squares, F Value = calculated F value, Adj Pr > F = Greenhouse-Geisser adjusted F value.

Source of variation	DF	Type III SS	Mean square	F value	Pr>F
Treatment	1	0.000650	0.000650	8.170000	0.006100
Plot(Treatment)	5	0.000780	0.000160	1.960000	0.100300
Depth	2	0.000310	0.000150	1.910000	0.157700
Depth*Treatment	2	0.000027	0.000014	0.170000	0.843600
Error	52	0.0042 00	0.000080		

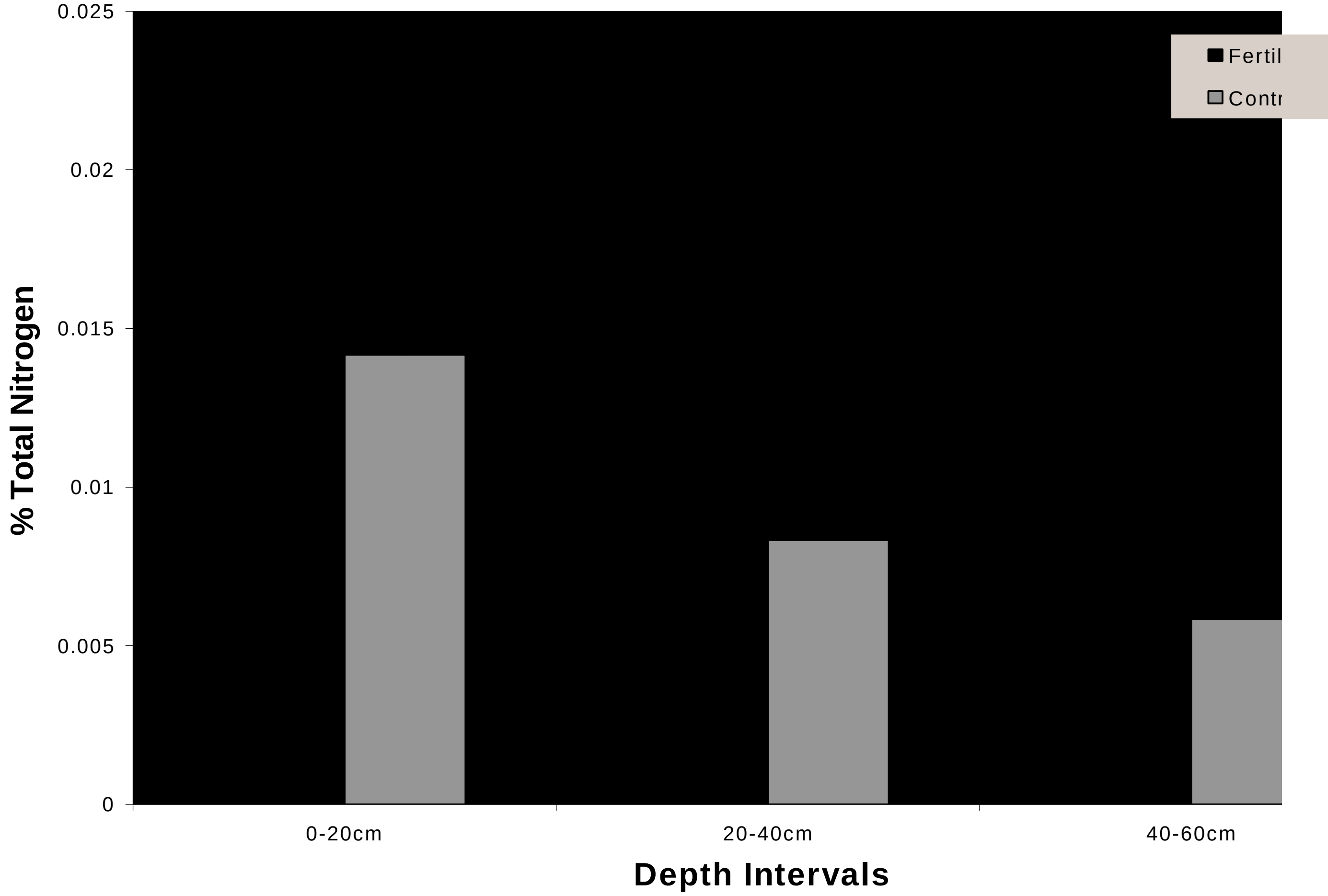


Fig. 9. Percent total nitrogen by mass for fertilized and control treatments for all three depth clas

DISCUSSION

Soil Nutrient Analysis

Unexpectedly, soil analyses did not show any significant differences in % total nitrogen by depth. However, Fig. 9 shows the high variability of these data. It is possible that a small sample size resulted in high variability that overwhelmed what should have been significant differences. In addition, the Carlo Erba analyzer may have been unable to precisely detect the low nitrogen levels inherent in sandy soils. This could also have lead to the high variability seen in these data. In another N-fertilization study at the same site, Day (1996) found greater nitrogen concentrations at shallower depths.

Root Length Elongation

The observed patterns of root length elongation rates were consistent with the body of literature that supports the hypothesis summarized by Hendricks et al. (1993) namely that fine root turnover rates increase with nitrogen availability. Root length elongation increased significantly due to the nitrogen fertilization regime. The soil analysis confirmed that % total nitrogen was significantly greater in the fertilized plots (Table 5) and there exists an extensive base of evidence that plants respond to fertilization through increased root production and biomass (Nadelhoffer et al. 1985, Safford 1974, Tillman and Wedin 1991). However, other researchers (Gower and Vitousek 1989, Vogt et al. 1987) have found conflicting results suggesting that fine root longevity and turnover rates are not significantly influenced by nitrogen availability.

The decrease in root length elongation with increasing depth was also expected. This is consistent with patterns of decreasing root elongation with depth found by a number of researchers in varied ecosystems (Hendrick and Pregitzer 1996, Weber and Day 1996) as well as decreasing total root biomass with increasing depth (Burke and Raynal 1994, Conn and Day 1993). Also, despite the results of the soil analysis, it is known that fertilizer placement tends to result in localizing the supply of nutrients in the upper portions of the soil (Drew and Saker 1975). A number of researchers have shown marked increases in the growth of lateral roots in response to localized nitrogen fertilization (Campbell and Grime 1989, Drew and Saker 1973, Gross et al. 1993, Hackett 1972).

While the effect of date is seemingly inconsequential ecologically, this finding is of some interest as the Tukey test showed only the first and the last date intervals to differ significantly. Precipitation for the year of the study was highest in May with a significant rainfall event occurring in the first interval of the study (Fig. 10). The stimulation effect of irrigation on root production is well established in the literature (Eissenstat and Yanai 1997). It is possible that the decrease in rainfall and the concurrent loss of moisture from the system, led to the significantly lower RLE values for the last interval of the study.

Mortality

The increase in mortality rates followed the expected trends in response to N fertilization. Aber et al. (1985) found increased turnover with nitrogen augmentation in a wide variety of ecosystems. Also, Gross et al. (1993) found increased mortality rates in fertilized soil patches. This result seems to support the hypothesis summarized in Eissenstat and Yanai (1997) for studies employing direct observation techniques that roots in nutrient stressed environments may benefit from longer lifespans and decreased turnover rates.

Mortality rates were consistent with expected results along the depth profile. Hendrick and Pregitzer (1996) and Joslin and Henderson (1987) found the highest mortality rates in the upper 20 cm of soil in forested ecosystems. As mentioned above, it is well established that the placement of fertilizer tends to result in localizing the supply of nutrients in the upper portions of the soil (Drew and Saker 1975). The results of this study would support the findings of Gross et al. (1993), showing a significant increase in root mortality in fertilized patches.

The effect of the depth*treatment interaction was interesting. The 22-46 cm depth class was affected more than the other two depth classes. This result suggests that, through time, increased N is effectively stimulating root turnover at progressively greater depths. It is possible that there is a system dependant limit to the effective depth of this phenomenon that should be examined in later studies. However, this result is not consistent with the long-term dataset. In fact, the two deeper depth classes seem to be more closely related throughout the long-term study (Day and Hutton, *unpublished data).*

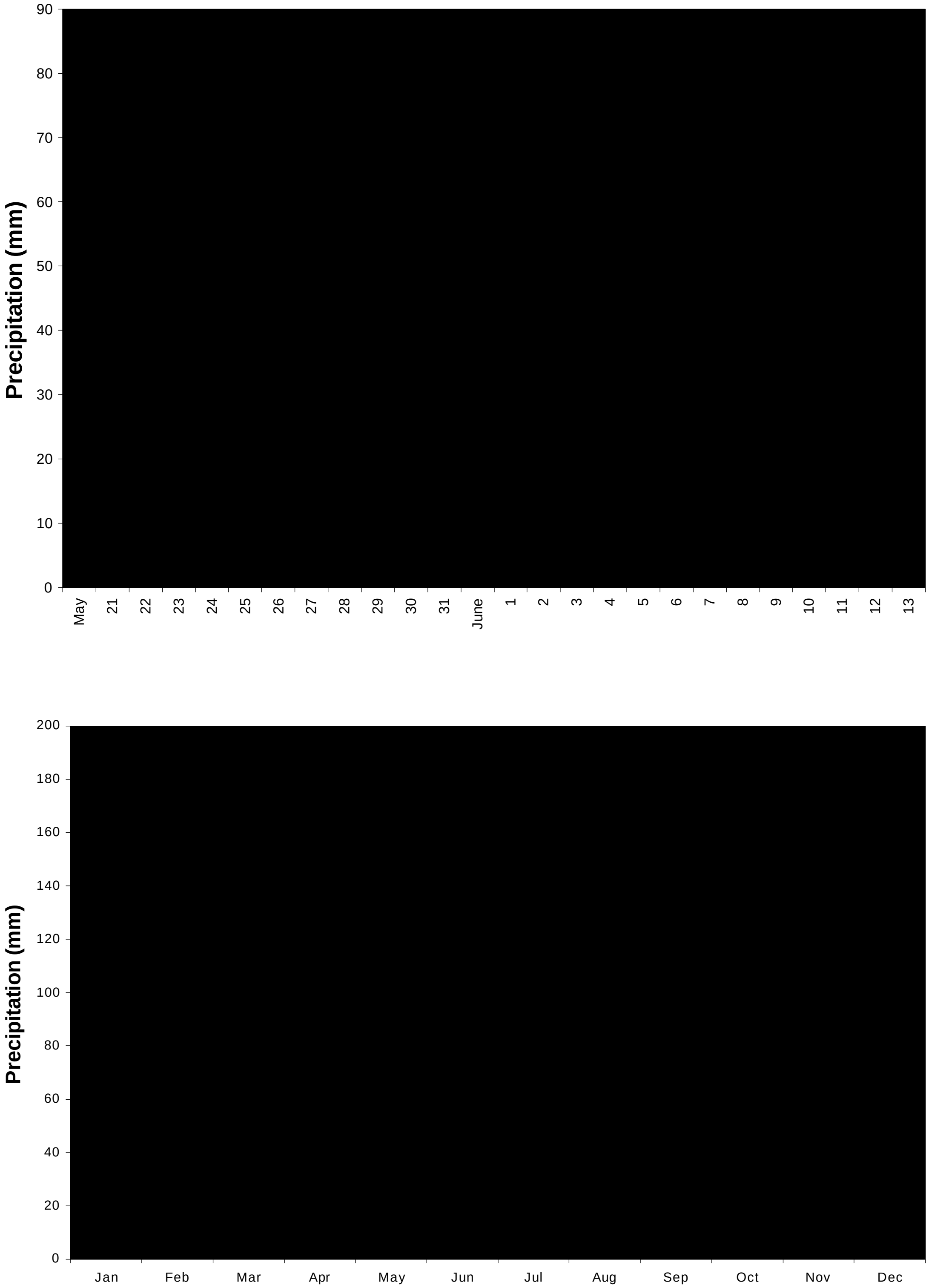


Fig. 10. Total daily rainfall for the period of 5/21/98 to 6/19/98 and total monthly rainfall for 1998 from the Hog Island weather station.

Root Length Elongation vs. Mortality

Mortality rates were consistently higher than root length elongation rates for the control group. This was surprising for this point in the growing season. The majority of belowground studies have shown positive production/mortality ratios for this period of the season in many varied ecosystem types (Burke and Raynal 1994, Hendrick and Pregitzer 1993a). However, even more surprising was the magnitude of stimulation in mortality rates due to N augmentation. The effect of N fertilization was much more pronounced on mortality rates than on root length elongation rates. This is suggestive of a belowground die back in response to nitrogen fertilization, perhaps associated with a shift in allocation to aboveground production.

Caution must be taken in interpreting these data. This finding may be more an artifact of the methodology employed. As mentioned earlier, roots were selected non-randomly from frames based on the presence of visible root tips. This was done under the assumption that only true root growth and/or dieback would be reflected by measuring these roots and any other root tips that entered the frames upon subsequent tapings. However, this may have biased the experiment resulting in this unexpected finding.

It is interesting that this trend was not reflected in the long-term dataset covering this whole growing season (Day and Hutton, *unpublished data*). In fact, root length density for the control group actually increased. This discrepancy may be due to the fact that the long-term dataset represents all roots within frames, whereas only select roots were analyzed in the short-term study.

The plots used in this study had been subjected to a N fertilization regime for 6 years prior to the beginning of this study. At this point, fine root dynamics would, presumably, be quite different from those at the onset of release from N availability constraints. Further studies need to examine fine root dynamics on a short temporal scale in conjunction with the initial fertilization event.

CONCLUSION

The results of this study support the second of the two hypotheses set forth by Hendricks et al. (1993a), stating that, in response to nitrogen fertilization, relative fine root carbon allocation remains constant and fine root turnover rates increase with nitrogen availability. Both fine root elongation and mortality responded to the nitrogen fertilization regime. However, the effect of nitrogen fertilization was much more pronounced on mortality rates than on root length elongation rates. This is suggestive of a below ground die back in response to nitrogen fertilization, perhaps associated with a shift in allocation to aboveground production. These findings are in contrast with the earlier findings of Weber and Day (1996) who observed decreased turnover with fertilization in the same plots. Further studies will be needed to elucidate the overall response of these nutrient limited plants to increased nitrogen availability.

LITERATURE CITED

Aber J D, Mellilo J M, Nadelhoffer K J, McClaugherty C A and Pastor J 1985 Fine root turnover in forest ecosystems in response to quantity and form of nitrogen availability: a comparison of two methods. Oecologia 66, 317-321.

Arthur M A and Fahey T J 1992 Biomass and nutrients in an Engelmann spruce subalpine fir forest in north central Colorado: pools, annual production, and internal cycling. Can. J. For. Res. 22, 315-325.

Birk E M and Vitousek P M 1986 Nitrogen availability and nitrogen use efficiency in loblolly pine stands. Ecology 67, 69-79.

Bohm W 1979 Methods of studying root systems. Ecological studies 33. Springer, Berlin, Germany.

Bowman W D,. Theodose T A, Schardt J C and Conant R T 1993 Constraints of nutrient availability on primary production in two alpine tundra communities. Ecology 74, 2085-2097.

Burke M K and Raynal D J 1994 Fine root growth phenology, production and turnover in a northern hardwood forest ecosystem. Plant and Soil 162, 135-146.

Campbell B D and Grime J P 1989 A comparitive study of plant responsiveness to the duration of episodes of mineral nutrient enrichment. New Phytol 112, 261-267.

Center for Remote Sensing 1989 ROOTS version 8-22-89. Michigan State University, East Lansing, Michigan, USA.

Chapin F S III 1980 The mineral nutrition of wild plants. Ann. Rev. Ecol. Syst. 11, 233-260.

Cheng W, Coleman D C and Box Jr J E 1991 Measuring root turnover using the minirhizotron technique. Agriculture, Ecosystems and Environment 34, 261-267.

Conn C E and Day F P 1993 Belowground biomass patterns on a coastal barrier island in Virginia. Bull. Torrey Bot. Club 120, 121-127.

Cutter G and Radford-Knoery J 1991 Determination of carbon, nitrogen, sulfur, and inorganic sulfur species in marine particles. Geophysical Monograph 63, 57-63.

Dalhman R C 1968 Root productivity and turnover of carbon in the root-soil matrix of a grassland ecosystem. *In*Methods of productivity studies in root systems and rhizosphere organisms. Ed. Ghilarov M S. pp 11-21. Nauka, Leningrad.

Day F P 1996 Effects of nitrogen availability on plant biomass along a barrier island dune chronosequence. Castanea 61, 369-381.

Drew M C and Saker L R 1975 Nutrient supply and the growth of the seminal root system in barley. Journal of Experimental Botany 26, 79-90.

Drew M C, Saker L R and Ashley T W 1973 Nutrient supply and the growth of the seminal root system in barley. Journal of Experimental Botany 24, 1189-1202.

Dueser R D, Graham M A, Hennessy G J, McCaffrey C, Niederoda A W, Rice T W and Williams B 1976 Ecosystem description: the Virginia Coast Reserve study. The Nature Conservancy, Arlington, Virginia, USA.

Ehrenfeld J G 1990 Dynamics and processes of barrier island vegetation. Reviews in Aquatic Sciences 2, 437-480.

Fogel R 1985 Roots as primary producers in below-ground ecosystems. *In* Ecological Interactions in Soil. Eds Fitter A H, Atkinson D, Read D J and Usher M. pp 23-36. Blackwell Scientific, Oxford.

Gleeson S K and Tillman D 1990 Allocation and the transient dynamics of succession on poor soils. Ecology 71, 1144-1155.

Grier C C, Vogt K A, Keyes M R and Edmonds R L 1981 Biomass distribution and above- and below-ground production in young and mature *Abies amabilis* zone ecosystems of the western cascades. Can. J. For. Res. 11, 155-167.

Gross K L, Peters A and Pregitzer K 1993 Fine root growth and demographic responses to nutrient patches in four old-field plant species. Oecologia 95, 61-64.

Hackett C 1972 A method of applying nutrients locally to roots under controlled conditions, and some morphological effects of locally applied nitrate on the branching of wheat roots. Aust. J. Biol. Sci. 25, 1169-1180.

Hayden B P, Dueser R D, Callahan J T and Shugart H H 1991 Long-term research at the Virginia Coast Reserve. Bioscience 41, 310-318.

Hendrick R L and Pregitzer K S 1992 The demography of fine roots in a northern hardwood forest. Ecology 73, 1094-1104.

Hendrick R L and Pregitzer K S 1993a The dynamics of fine root length, biomass and nitrogen content in two northern hardwood forests. Canadian Journal of Forestry Research 23, 2507-2520.

Hendrick R L and Pregitzer K S 1993b Patterns of fine root mortality in two sugar maple forests. Nature 361, 59-61.

Hendrick R L and Pregitzer K S 1996 Temporal and depth-related patterns of fine root dynamics in northern hardwood forests. Journal of Ecology 84, 167-176.

Hendricks J, Nadelhoffer K J and Aber J D 1993 Assessing the role of fine roots in carbon and nutrient cycling. Tree 8(5), 174-178.

Joslin J D and Henderson G S 1987 Organic matter and nutrients associated with fine root turnover in a white oak stand. Forest Science 33, 330-346.

Kachi N and Hirose T 1983 Limiting nutrients for plant growth in coastal sand dune soils. Journal of Ecology 71, 937-944.

McMichael B L and Taylor H M 1987 Applications and limitations of rhizotrons and minirhizotrons. Minirhizotron observation tubes: methods and applications for measuring rhizosphere dynamics. *In* American Society of Agronomy Special Publication Number 50. Ed Taylor H M pp 1-13. Madison, Wisconsin, USA.

Nadelhoffer K J, Aber J D and Melillo J M 1985 Fine roots, net primary production, and soil nitrogen availability: a new hypothesis. Ecology 63, 1481-1490.

Neill C 1992 Comparison of soil coring and ingrowth methods for measuring belowground production. Ecology 73, 1918-1921.

Powell S W and Day F P 1991 Root production in four communities in the great dismal swamp. American Journal of Botany 78, 288-297.

Pregitzer K S, Hendrick R L and Fogel R 1993 The demography of fine roots in response to patches of water and nitrogen. New Phytologist 125, 575-580.

Raich J W and Nadelhoffer K J 1989 Belowground carbon allocation in forest ecosystems: global trends. Ecology 70, 1346-1354.

SAS Institute Inc. 1985 SAS User’s Guide: Statistics. Cary, North Carolina, USA.

Safford L O 1974 Effects of fertilization on biomass and nutrient content of fine roots in a beech-birch-maple stand. Plant and Soil 40, 349-363.

Sharpe P J H abd Rykiel E J 1991 Modeling integrated response of plants to multiple stresses. *In* Response of plants to multiple stresses. Eds. Mooney H, Winner W and Pell E pp 205-224. Academic Press, San Diego, California, USA.

Stevenson M and Day F P 1996 Fine-root biomass distribution and production along a barrier island chronosequence. Am. Midl. Nat. 135, 205-217.

Tilman D and Wedin D 1991 Plant traits and resource reduction for five grasses growing on a nitrogen gradient. Ecology 72, 685-700.

Upchurch D R and Ritchie J T 1983 Root observations using a video recording system in mini-rhizotrons. Agronomy Journal 75, 1009-1015.

Van der Vak A G 1974. Mineral cycling in coastal foredune plant communities in Cape Hatteras National Seashore. Ecology 55, 1349-1358.

Verardo D J, Froelich P N and McIntyre A 1990. Determination of organic carbon and nitrogen in marine sediments using the Carlo Erba NA-1500 Analyzer. Deep-Sea Res. 37, 157-165.

Vitousek P M and Sanford R L 1986 Nutrient cycling in a moist tropical forest. Ann. Rev. Ecol. Syst. 17, 137-167.

Vogt K A, Grier C C and Vogt D J 1986 Production, turnover, and nutrient dynamics of above- and below-ground detritus of world forests. Adv. Ecol. Res. 15, 303-307.

Weber E P and Day F P 1996 The effect of nitrogen fertilization on the phenology of roots in a barrier island sand dune community. Plant and Soil 182, 139-148.

Wells B W and Shunk . V 1938 Salt spray: an important factor in coastal ecology. Bull. Torrey Bot. Club 65, 485-503.

Willis A J 1965 The influence of mineral nutrients on the growth of *Ammophila arenaria*. Journal of Ecology 53, 735-745.

Wilson S D and Tilman D 1991 Components of plant competition along an experimental gradient of nitrogen availability. Ecology 72, 1050-1065.

VITA

John Hutton was born in Aberdeen, MD. He attended grade and high school in Chattanooga, TN. At the University of Georgia, he majored in Botany. He also worked on an ecological research project, funded by the National Science Foundation, at Coweeta Hydrological Laboratory. At Old Dominion University, he worked with Dr. Frank Day, studying fine root response to nitrogen fertilization. John Hutton currently resides in Atlanta, GA where he works as a Wetlands Ecologist for the Georgia Department of Transportation.