THE EFFECT OF NITROGEN FERTILIZATION ON THE PHENOLOGY OF ROOTS IN A BARRIER ISLAND SAND DUNE COMMUNITY

by

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Abstract

Little work has been done on the phenology of root growth and senescence largely due to methodological difficulties. The application of minirhizotron technology has enaWed the tracking of individual roots through an entire growing season. As a result, direct measures of turnover, root growth, and an analysis of cohorts were made. Small plots on a 36 year old dune on Hog Island, a barrier island in the Virginia Coast Reserve Long Term Ecological Research Site, were fertilized with nitrogen. Minirhizotron tubes were installed in each fertilized and control plot. Each tube was sampled monthly for nine months, March through October of 1992. Root length density increased throughout the growing season with the highest root length density in the top 0-20 cm of the soil profile. The fertilized plots had higher root length densities (14.05 mm cm^{-2}) than the unfertlized plots (2.68 mm cm^{-2}). The turnover was higher in the unfertilized plots only in the top 0-20 cm of the soil profile (fertilized = 0.020 percent loss per day, unfertilized 0.024 percent loss per day). The cohort analysis found that the largest loss of roots for a cohort occurs within the first month. There was also a decline in root loss in the last sampling of the last cohort potentially indicating the roots were preparing ‘for the winter months. The overall low turnover rate, the decreased turnover rate with fertilization and the decreased turnover in the last cohort imply that roots tend to be conserved in this nutrient poor system.

Keywords: Root, Minirhizotrdn, Phenology, Turnover, Nitrogen Fertilization

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Introduction

Plants primarly acquire nutrients through root systems. Roots are also the largest heterotrophic portion of the plant. As a result, a balance must be reached between the needs of the plant for nutrients end the considerable energy drain thet roots place on the plant (Caldwell 1979, Bloom et al. 1985). Little work hes been done to explore the root distribution patterns that plants use belowground to survive in nutrient limited ecosystems.- Barrier islands provide a particularly good environment to observe, nutrient limited root distribution responses. The sandy soil of the islands makes root observetion relatively easy end the low nitrogen status of the soil also makes nitrogen content of the soil easily manipulated.

Roots serve four major roles for most plants : holding the plant in the soil, resource storage, absorbing nutrients, end absorbing water. Caldwell (1979) described the high expense of root growth end maintenance end the significant resource drain placed on the plant. Because roots are an expensive portion of the plant to grow end maintain, plants heve developed strategic ways to distribute their roots. Harris end Wilson (1970) found that four grasses showed different strategies of root development and that the strategy's effectiveness wes directly related to the severity of stress placed upon the plant. The position of roots within the soil metrix should, therefore, reflect the strategies plants use to efficiently perform root functions.

Plants have evolved strategies for efficiently managing environmental stresses (Bloom et al. 1985). Caldwell (1979) suggested that short lived fine roots placed within the established root zone would be more effective at gathering ephemeral resources from microsites within the soil matrix than would long lived roots. He also suggested that nutrient adsorptive intensity may be inversely related to root longevity. Both Grime and Cambell (1991) and Sharpe and Rykiel (1991) described' productivity responses to resource availability within a site. They felt that highly productive, resource rich plants have flexible allocation patterns while resource poor, unproductive plants have a less flexible response. Sharpe and Rykiel (1991) also suggested that resource poor plants tend to allocate resources to storage when encountering a new supply, while Grime and Cambell (1991) detailed on a cellular level how this response would occur. Tilman and Wedin (1991) found an increase in root biomass correlated with increasing nitrogen status for five perennial grass species.

Previous research on roots has focused mostly on root/shoot ratios (Mooney and Winner, 1991; Bloom et al. 1986; Thornley, 1969; Orians and Solbrig, 1977; Hansson and Petterson, 1989; Bazzaz et al., 1987); however, little work has been done on the distribution of roots within the soil matrix after root/shoot partitioning has occurred. There has also been little work done on root length density variation with depth and time, and no studies have been performed in dune communities.

Turnover has also received little attention in dune communities. Aber et al. (1985) found turnover directly correlated with an increasing nutrient status in a forested ecosystem, which supports both Grime and Cambell's (1991) and Sharp8 et al.'s (1991) theory on resource allocation.

Traditional root research involves such time-consuming and destructive methods as soil coring or soil monolith removal. The destructive nature of traditional sampling makes repeated measurements of the same volume of soil impossible. Repeated coring also causes a significant impact on the research site. Current methods of turnover calculations through sequential corings are also known to be inaccurate (Singe et al. 1984). Minirhizotrons, clear tubes in which video tape recordings of roots are made, have been used at a variety of other sites including a hardwood forest (Hendrick and Pregitzer 1992a, Hendrick and Pregitzer 1993a, Hendrick and Pregitzer 19936) agricultural systems (Hansson and Andren 1987) and a simulated tropical forest (Körner and Arnone 1991). Minirhizotrons were used to observe root dynamics in the present study because their non-destructive nature allows direct measurement of turnover and root longevity. Because there is little site disturbance after the installation of the minirhizotrons, there is much less disturbance to the experimental plots. Repeated measurements can be made of the same section of soil and direct measurements of root longevity and turnover are obtained by measuring the same roots through time.

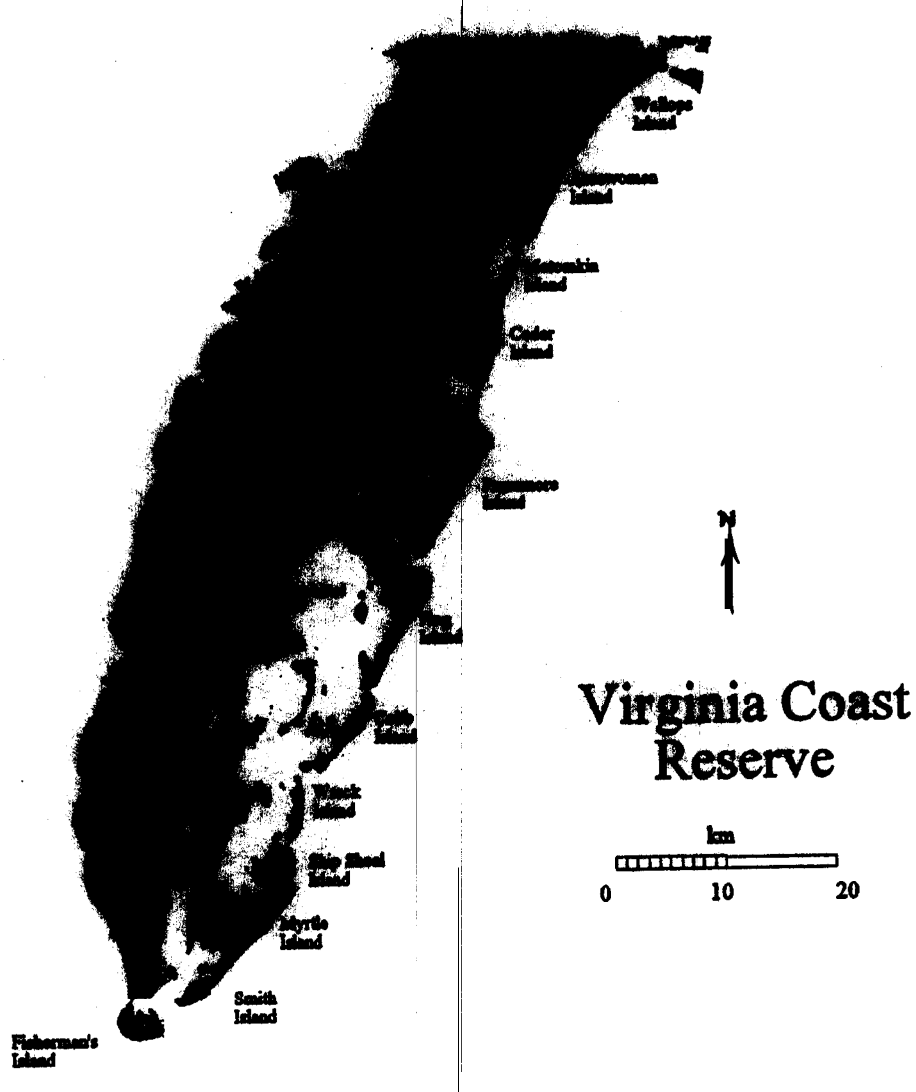
This study quantified the root dynamics of a barrier island dune community to determine what strategies are used by the plants to survive. The dune ecosystem on which the study site is located is a low productivity, low resource environment and the plants, primarily perennial grasses, should respond as previously described for low resource, low productivity plants. In addition, the location of nutrients within the soil profile should affect the location of the roots. There should be an increase in longevity of roots within resource rich patches, and a decrease in longevity in nutrient poor patches. The primary questions posed in this study included: Does turnover increase with nitrogen fertilization? Do ephemeral roots exist and does fertilization affect their longevity? Is there an increase in root length density which coincides with fertilization within the soil profile? Is there a seasonal change of turnover? Is there a seasonal change of root (length density? The primary objective of this study was to quantify root phenology, as expressed in root length denslty and turnover, in fertilized and control plots on a nutrient poor barrier island sand dune using minirhizotron observation tubes.

METHODS

Site Description

The study site is located on Hog Island, a barrier island off the eastern coast of the Delmarva Peninsula on the Virginia Coast Reserve (VCR) Long-Term Ecological Research (LTER) Site (Figure 1). The land is currently owned and managed by the Nature Conservancy of Virginia, located at Brownsville near Nassawadox. Although a small community existed on Hog Island in the

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


Wallops Island
Cedar Island
Wreck Island
Ship Shoal Island
Myrtle Island
Smith Island
Fisherman's Island
N
Virginia Coast Reserve
km
0
10
20

early part of this century, the island has been largely uninhabited since the late 1940's (Dueser et al, 1976). All feral domestic livestock were removed from the island in the cattle drive of 1980 (Hayden et al. 1991).

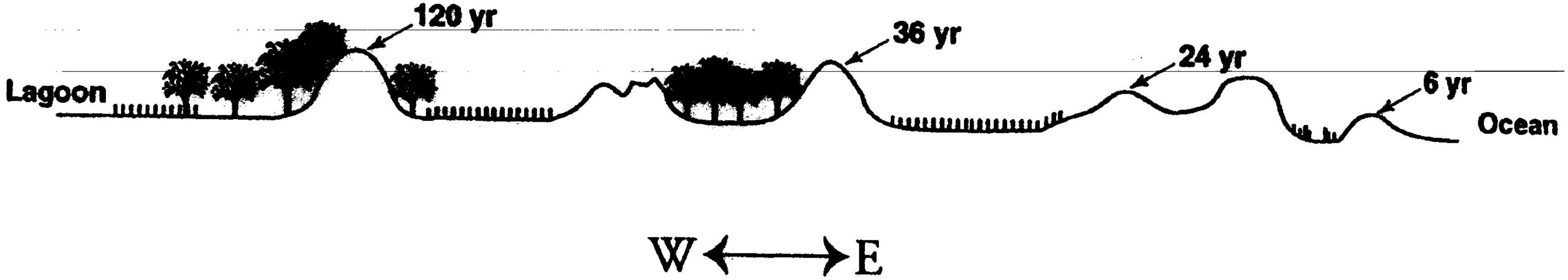
On north Hog Island accretion has produced distinct dune complexes as well as a foredune area. From the Atlantic Ocean to the bayside of the island, a chronosequence of dunes have been aged. from 6 to 124 years old (Hayden et al, 1991). The present study was located on the 36 year old dune ridge (Figure 2).

The plant community on this dune complex is dominated by *Ammophila breviligulata* Fernald, *Spartina patens* Muhl. and *Panicum amarum* Eli. (Table 1). The community is on a well-drained sand dune with surrounding wet areas. To the east Is a freshwater marsh of *Spartina patens*. To the west are wax myrtle (*Myrica cerifera* L.) thickets.

Monthly rainfall and temperature data for 1992 ranged from highs of 216 mm rainfall (August) and 25.1 °C mean temperature (July) to lows of 5.7 mm rainfall (December) and 0°C mean temperature (December) (Table2).

The soil of the study site is a Newhan-Corrolan complex (Dueser et al, 1976). This udipsamment is characterized by excessively drainedNewhan soil in the higher elevations and the well drained Corrolan soil in the lower elevations. The soil, therefore, provides few nutrients, low nutrient retention and limited water retention. All plots were placed within the Newhan series. A previous study (Day and Lakshmi, unpublished data) found a higher level of

Figure 2. Diagram of dune chronosequence on Hog Island; study plots are on the 36 year old dune.



120 yr
36 yr
24 yr
6 yr
Lagoon
Ocean
W ⟷ E

Table 1. Plant species in both fertilized and unfertilized plots ranked by mean cover class (data from two quarter meter square samples in each plot sampled 9/94). Ambr = *Ammophila breviligulata* Fernald, Paam = *Panicum amarum* Ell., Sppa = *Spartina patens* Muhl., Soca = *Solanum carolinense* L., Ansc = *Andropogon scoparius* Michaux, Livi = *Linum virginianum* L., Ruac = *Rumex acetosella* L., Sote = *Solidago tenuifolia* Pursh, Casp = *Carduus sp.*

Fertilized Plots			Unfertilized Plots		
Species	Stems m^{-2}	% Cover Class'	Species	Stem m^{-2}	% Cover Class'
Ambr	315	2.375	Sppa	82	1
Paam	39.5	1.125	Ambr	34	0.625
Sppa	10.5	0.5	Paam	9.5	0.375
Soca	2.5	0.375	Soca	0.5	0.25
Ansc	6	0.125	Ruac	13	0.25
Livi	0.5	0.125	Casp	0.5	0.125
Ruac	0.5	0.125	Ansc	0	0
sote	1	0.25	Livi	0	0
Casp	0	0	Sote	0	0

'Mean % Cover Classes (4 = 75-100%, 3 = 50-75%, 2 = 25-50%, 1 = 1-25%, 0 = 0%)

Table 2. Average monthly temperature and total monthly rainfall from the Hog Island weather station in 1992.[a]

Month	Temperature (°C) /Rainfall (mm)
Jananuary	4.9 / 40.8
Febuary	5.6 / 68.9
March	6.7 / 40.2
April	1 2.2/45.5[b]
May	15.1/131.9[b]
June	20.5 / 107.8
July	25.1 / 102.7
August	23.2 / 216.0
September	21.9 / 162.8
October	14.7 I 44.6
November	11.1 / 101.0
December	0.0/5.7[b]

[a]Data condensed from Krovetz and Porter (1992)
[b]Data from Brownseville Virginia to supliment missing weather data from Hog Island

soil water nitrogen at 15 cm than at 50 cm in areas with the same fertilization treatment used in this study and in areas without fertilization (Table 3).

Experimental Design

Eight 3x3 m plots were selected within the study site. The sites were chosen for both visually representing the dune ecosystem and being spaced far enough apart to allow for independent treatment of the plots. Four plots were randomly assigned to nitrogen fertilization treatment and four plots were left as untreated controls. Fifteen g N m^{-2} with a 70%-30% mix of coated temperature-release urea to uncoated urea was applied to the fertilized plots. There were three applications of fertilizer during the year (March 14, June 10, and October 3 of 1992). The site was monitored on an approximately monthly basis from March through October of 1992 (Table 4). Inclement weather and logistical problems prevented strict monthly sampling. Four minirhirotron tubes were placed in each plot, one meter from the adjacent sides, each tube perpendicular to one of the sides (Figure 3). The etched frames faced towards the center of the plot to reduce any possible edge effects,.

Ail eight 3x3 m plots were chosen within the study site to reflect the grassy drier regions of the dunes. The plots were, therefore, away from *Myrica* thickets and above any areas which appeared to have experienced standing water. in addition, the plots were distributed in such a way to assure that the fertilization treatment of any single plot would not affect an adjacent plot.

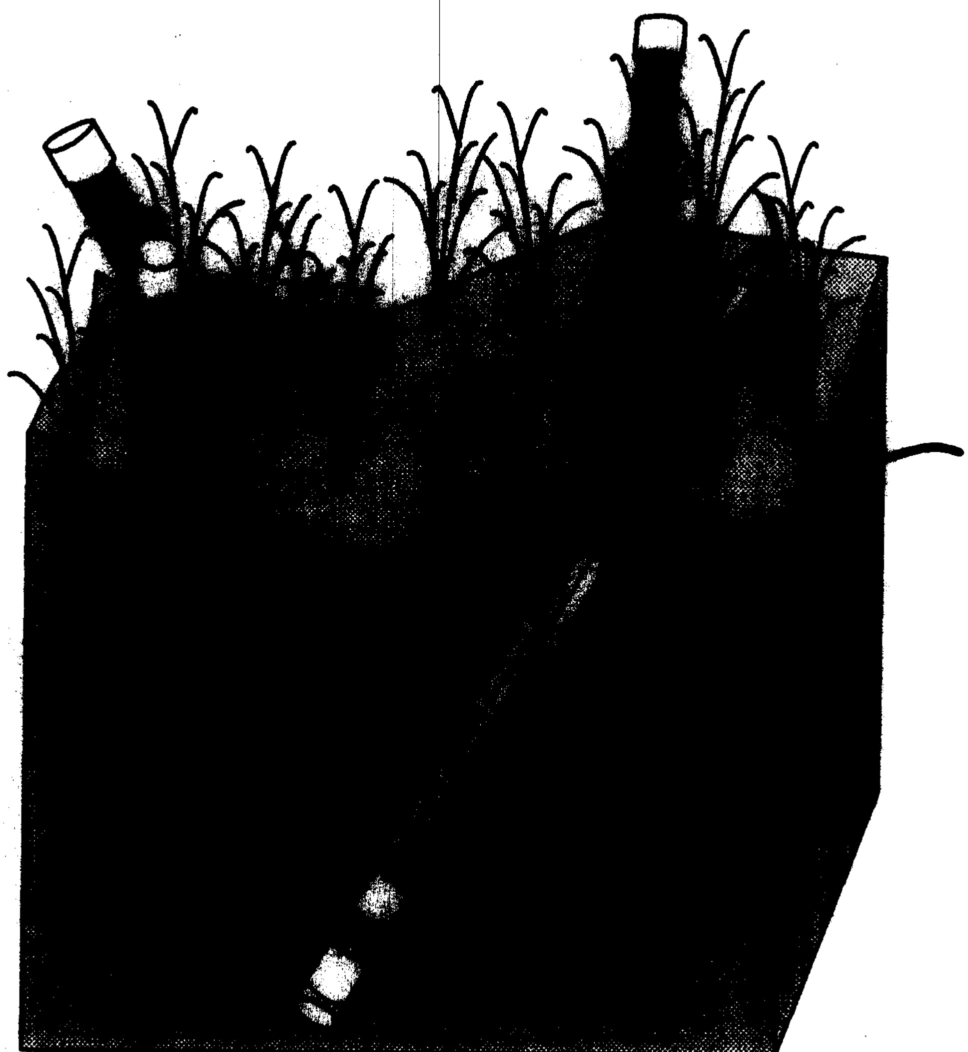
Table 3. Ppm ammonia and nitrate from porous cup lysimeters at two depths on the 36 year old dwne on Hog Island(Day and Laksmi, unpublished data)

	Control		Fertilized	
depth	Ammonia	Nitrate	Ammonia	Nitrate
15 cm	0.08	0.29	19.86	29.28
50 cm	0.05	0.07	0.36	18.16
Ratio	1.6	4.14	55.2	1.6

Table 4. Dates that observations were made from research site during 1992.

March 14,1992
April 25,1992
May 28,1992
June 24,1992
July 29,1992
August 25,1992
October 3,1992
October 24,1992

Figure 3. Installation of tubes within a plot. A minirhizotron camera is inserted in one of the tubes.



Minirhizotrons

Minirhizotrons are clear tubes placed in the ground from which observations are made of the roots in the surrounding soil. In the present study, observations were made with the video camera system described by Hendrick and Pregitzer (1992s).

Minirhizotron research requires three main steps: field installation and data collection, laboratory analysis/ and data processing. The first step involves preparing, installing, and monitoring the tubes. Laboratory analysis entails the creation of data sets from video tapes made in the field. Finally, the data processing provides ecologically significant information such as root length density, turnover rate, and phenology of cohort groups.

The minirhizotron tubes were 2 m long, clear, 5.08 cm inner diameter butyrate tubes with 0.65 cm thick watts. Butyrate tubes have been used by several other researchers (Hendrick and Pregitzer 1992*a*, Rygiewicz et al., 1991) and are more durable than glass tubes (Hendrick, pers comm.).

The bottom of each tube was capped with plexiglass to prevent soil and water from entering, and thu [illegible] vent the accumulation of fungi, algae, or samplings. Two parallel lines, 18 mm apart, were etched down the surface of the tube, from the top to the bottom, with transverse lines etched every 13.5 mm. The result was a column of frames or "windows", 18 mm x 13.5 mm, stacked from the base to the top of the tube. A number was placed in the third frame from the bottom of each tube to provide unique identification.

Etch marks were filled with acrylic fluorescent green paint, as suggested by Pregitzer (1992). Thinned paint was brushed into the etchmarks and after partial drying, excess paint was wiped off with a damp cloth. This produced clear windows surrounded by fluorescent green lines.

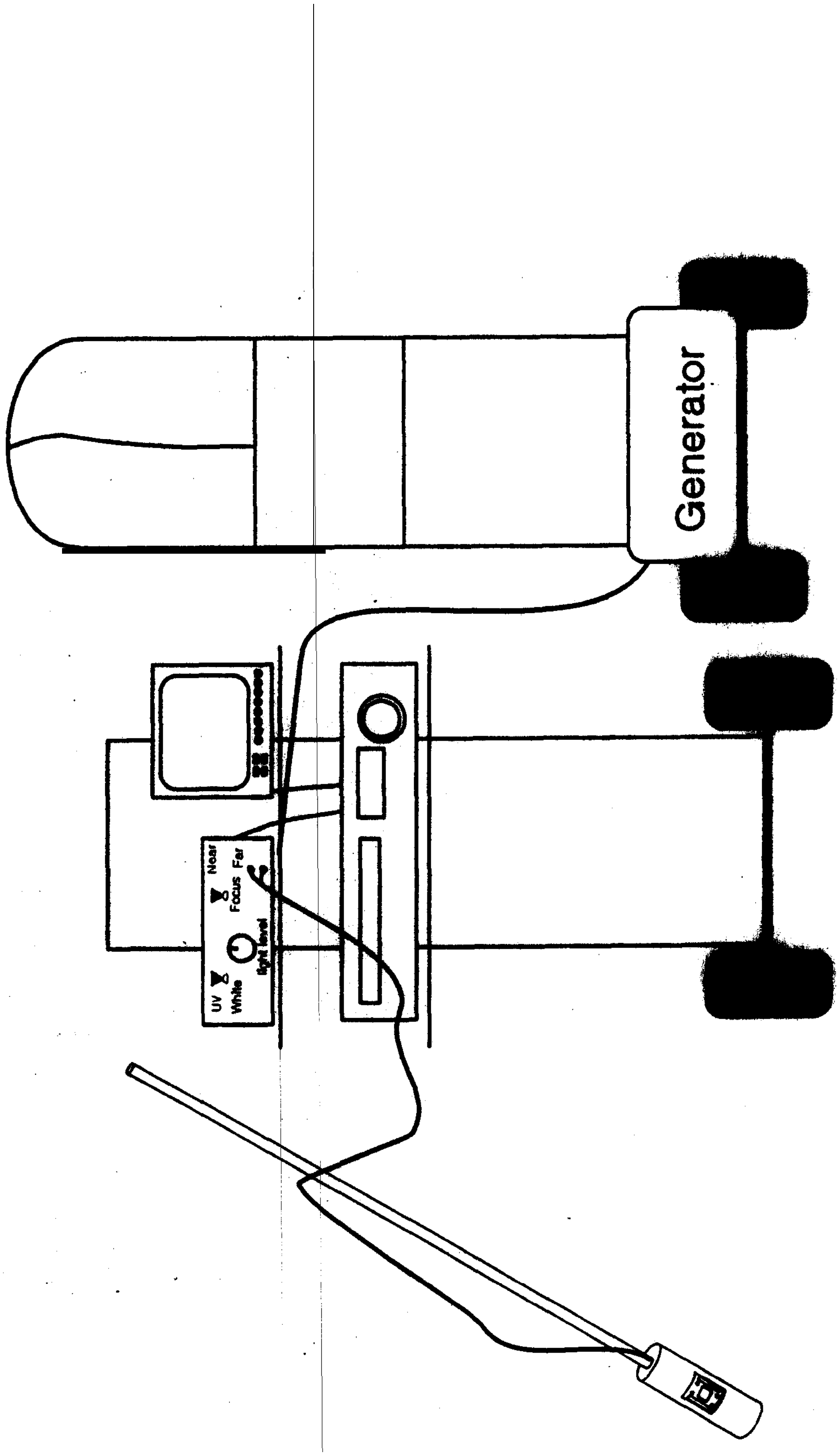
Light has been shown to Influence root growth (Lake and Slack 1961, Furuya and Torrey 1964) and several other studies have shown the effect of light on growth of roots along minirhizotron tubes (Levan et al. 1987, Vos and Groenwald 1987). To prevent light from penetrating the minirhirotron tubes, the top of each tube was painted to just below the soil surface with a flat black paint. In addition, the top of the tube was wrapped with electrical tape to provide a light-tight fitting with a PVC cap which was placed over the open end of the tube. The cap also prevented debris and rain from entering the tube.

Each tube was inserted into the soil at a 45° angle to the ground. The angle of insertion was assured with an auger stand built at the Old Dominion University science shop. Holes were augered to a depth which would allow observation through approximately 80 frames or 1 m of observable tube. After the depth was estimated to be correct for insertion of the tube, the tube was inserted in the augered hole. The top was then wrapped with tape and capped. Loose sand was replaced around the interface of the tube and the soil-surface. The tubes were installed Febuary 15-29, 1992.

Once the tubes had been in place for two to three weeks, the first readings were taken. A Bartz two-inch diameter minirhitotron video camera was inserted into the minirhizotron tubes. The camera was placed at the bottom of each tube and drawn along the etched marks pausing at each window. The image was viewed on a small color monitor and focused with an electronic control box while recording in S-VWS on a VCR (Figure 4). The tapes were dated and returned to the laboratory for analysls.

In the laboratory, the same VCR used in the field was hooked to a Targa plus video board in the computer. This board converts the signal from an analog television signal to a digital signal which can be interpreted by a computer monitor. The board also stores the images, frame by frame, in memory; thus, video frames were "frozen" for analysis. ROOTS software ver "1 (Michigan State University Remote Sensing Laboratory, 1989) was used to digitize the video images from the field to numeric data. A computer "mouse" was used to outline a skeleton (a line along the center and a single line along the width) of the root on the monitor and to store the coordinates, as well as length and width measurements, to a dbase file (see Appendix for detailed protocol). The root skeletons ofr m the previous month were overlaid on the corresponding video frame from the current month. Whether the roots within

Figure 4. Illustration of minirhizotron field equipment **which includes: minirhizotron camera, minirhizotron** camera controller, super **VHS VCR, monitor, and generator.**


Generator
UV
White
light level
Focus
Near
Far

the current video frame matched that of a previous root skeleton was noted. After digitizing was complete, the files created by ROOTS were compiled into a single file whii was manipulated in Microsoft Foxpro (a database program).

Within Foxpro, programs were developed which produce cohort groups and turnover data. Because black roots were shown to produce new live roots, color was not useful for determining root longevity in this study. The program therefore assumed all visible roots were alive. The database programs placed each root into a cohort based on its first appearance (programs and required digitizing protocol available upon request from the author). The cohort group provided the basis for turnover calculations. The data were exported to an ASCII file and loaded into SAS for statistical analyses.

Root Length Density

Root length density (RLD) was calculated by summing all of the foot lengths (RL) for a given area (A) of the tube and dividing that quantity by the area of the tube observed. Root length density was calculated for each date and depth combination of each tube for analysis.

Turnover

To calculate turnover, the root length was compared between sample n and sample n+ 1. If a root was not observed at time n + 1 the root was assumed to have decomposed. The turnover was therefore 1.00 or 100%. If the root was longer at time n + 1 than at time n, in other words the root grew between time n and n + 1, there was no turnover and the turnover was 0.00.

If a root was smaller at n + 1 than at n, a simple calculation was made to determine the turnover (equation 1).

(1) (root length n - root length n + 1) / root length n = turnover

This measure was related to ndrick and Pregitter's (19928) mortality measure butbecause it was fig red on a per root basis, with root extension eliminated from the calculation, tr e turnover was actually measured. Turnover was calculated on a per root basis and averaged for each depth and date combination of each tube. These means were then used for statistical analyses.

Cohort Analysis

To determine root life expectancy, roots were placed into cohorts, or groups, based upon when the roots were initially observed. Percent change was calculated from time n to time n+ 1 for each cohort. The percent change was then divided by the number of days between sampling dates to control for different intervals between sampling dates. Root number density rather than root length density was used. This eliminated skewing. the data in favor of longer roots.

Not all of the data were used for the statistical analyses. The reasoning for the removal process folk)ws. It was believed that the largest percent change would occur between the irst and second month of any cohort. Three consecutive percent changes were felt to be the minimum to ensure that the changes observed were not transitory. Four months of data were, therefore, determined to be the minimum number of sampling dates within any cohort. The first, second, third, fourth, and fifth cohorts had enough sampled dates to meet the minimum criteria to be chosen for this analysis. The first two cohorts were eliminated from the analysis because they contained too many missing values (percent change could not be calculated if there were no roots observed within a cohort). Therefore, only the third, fourth, and fifth cohorts were used in the analysis.

Because interest was focused upon the longevity of the roots, the analysis used root age. Root age was given by the number of dates since the roots were first observed, as discussed above. There were three root age categories for the percent change: one, two, and three. Root age one represented the percentage change interval from time one to time two. Root *age* two represented the percentage change interval from time two to time three and so forth. If a root lives to be a certain age, it *may* have a greater likelihood of survival. A percent change would, therefore, decrease with root age. Another possibility is that as roots age their likelihood of survival would decrease. This would result in higher percent changes with root age.

Statistical Analyses

Originally, it was hoped that tubes could be nested within plots as suggested by Hendrick and Pregitzer (19926): however, there were not enough degrees of freedom with the number of factors and levels measured and the number of replicates to calculate an error term for the ANOVA. Therefore, the option of which factors and/or' levels to decrease had to be decided. Originally frames were classed into 12 depth classes; each depth class was roughly equivalent to 5 cm vertical depth. Because there were not enough-degrees of freedom available to look at all of the depths separately, the depths were pooled into three roughly 20 cm vertical depth classes (O-22 cm, 22-46 cm, 46-68 cm). This provided enough degrees of freedom for the error term to perform the analyses. Because /depth class one was necessarily related to depth class two and depth class two was related to depth class three, depth was analyzed as a repeated measure. Dates were also analyzed as a repeated measure for this study. The final ANOVA model, therefore, had plots nested within treatments and both time and depth class as repeated measures.

A nested, crossed, repeated measures model wes used to analyse the cohort data. Plots were nested within treatments as was done with the other measures. However, cohorts were crossed with treatment and root age was a repeated measure.

Initial testing of the root length density data showed that a depth*date*plot(treat) interact on was significant. When the data were plotted depth*date for each plot, t was apparent that plot 2 was significantly different from all of the other plots. This plot, although an unfertilized plot, had the highest root length density of any depth and date combination for either the fertilized or the unfertilized plots. This plot also had a higher root length density in the 20-40 cm depth class than the O-20 cm depth class. All other plots had the highest root length density in the first O-20 cm depth class, nearest the surface. Both of these trends showed clear differences not only from the other unfertilized plots but from the fertilized plots as well. Since the plot was initiated in the spring of 1992, *Myrica cerifera* began to overhang the plot and possibly had roots extending into the dune edge. Thus, because plot 2 differed not only in the quantity of roots but also in the pattern of root distribution from all the other plots, it was considered an outlier. Because it clearly was not representitive of the dune community, plot 2 was removed from all analyses.

A log transformation was used in an attempt to normalize the root length density data. Biological data, such as root length density, are often distributed along a log-linear scale. Because the analysis was an unbalanced design (plot 2 was removed), a sum of squares IV was used. A test for sphericity of orthogonal components was found to be significant ($p < .0001$); therefore, the Greenhouse-Geiser adjusted F was used rather than using the split plot F probabilities. The Greenhouse-Geiser adjusted F deflates the degrees of freedom to compensate for the interrelatedness often found in repeated measures. It also adjusts to aptly measure the repeated factor. It takes into account the fact that time one is more highly correlated with time two than with time n and similarly for the depths.

Because turnover is a rate, an arcsine transformation was used in an attempt to normalize the data. Missing values reduced the degrees of freedom so that it was not possible to calaculate the error term for the ANOVA. The first three turnover intervals were removed from the analysis to remove enough missing values to allow the calculation of the error term; missing values were highest in the first few months of sampling. A test for sphericity of orthogonal components was not found to be significant ($p > 0.5943$). As a result, the split-plot F probability was used rather than the adjusted F used in the root length density analysis.

Results

Root Length Density

The results showed three main effects and two interactions to be significant (Table 5). Figure 5 shows root length density for all depths and dates for both the fertilized and unfertilized treatments. The same depth pattern of root length density can be seen in both treatments with the highest root length density occuring at ≈15 cm. The higher variation seen in the unfertilized plots can be attributed to the fine scale shown for root length density in the unfertilized graph than in the fertilized graph. Lower root numbers mean that fewer roots can provide information regarding root length density Both depth*date ($F=4.40, p<0.01$) and date*treatment ($F= 10.51, P<0.01$) interactions were significant. The fertilized treatment increased in root length density over time; whereas, the unfertilired treatment only increased slightly over time (Figure 6). As a result there was also a higher root length density in

Table 5. Nested doubly repeated measures analysis of variance examining the effect of fertilizer on root length density over time and across three depth classes. DF = Degrees of freedom, SS = sums of squares, F Value = calculated F value, Adj Pr > F = Greenhouse-Geisser adjusted F value.

Source of variation	DF	Type IV SS	Mean square	F value	Pr>F
Treat'	1	343.763	343.763	43.16	0.0001
Plot (Treat)[a]	5	125.385	25.077	3.15	0.0282
Error	21	167.246	7.964		
Depth"	2	310.933	155.467	30.71	0.0001
Depth *Treat	2	1.433	0.716	0.14	0.8680
Depth*Plot(Treat)	10	56.187	5.619	1.11	0.3776
Error(depth)[b]	42	212.626	5.063		
Date[a]	7	1043.13	149.019	69.1'8	0.0001
Date *Treat[a]	7	158.404	22.629	10.51	0.0001
Date*Plot(Treat)	35,	118.880	3.397	1.58	0.0529
Error(Date)[c]	147	316.646	2.154		
Depth *Date[a]	14	70.265	5.019	4.40	0.0001
Depth*Date*Treat	14	11.357	0.811	0.71	0.6688
Depth*Date*Plot(Treat)	70	112.903	1.613	1.42	0.0757
Error(Depth *Date)[d]	294	334.983	1.139		

[a] significant at the $p<0.05$ level, [b] Greenhouse-Geisser Epsilon = 0.9977,
[c] Greenhouse Geisser Epsilon= 0.7607, [d] Greenhouse Geisser Epsilon = 1.0664

Figure 5. Contour plot of root density by both depth and date for fertilized and unfertilized plots.

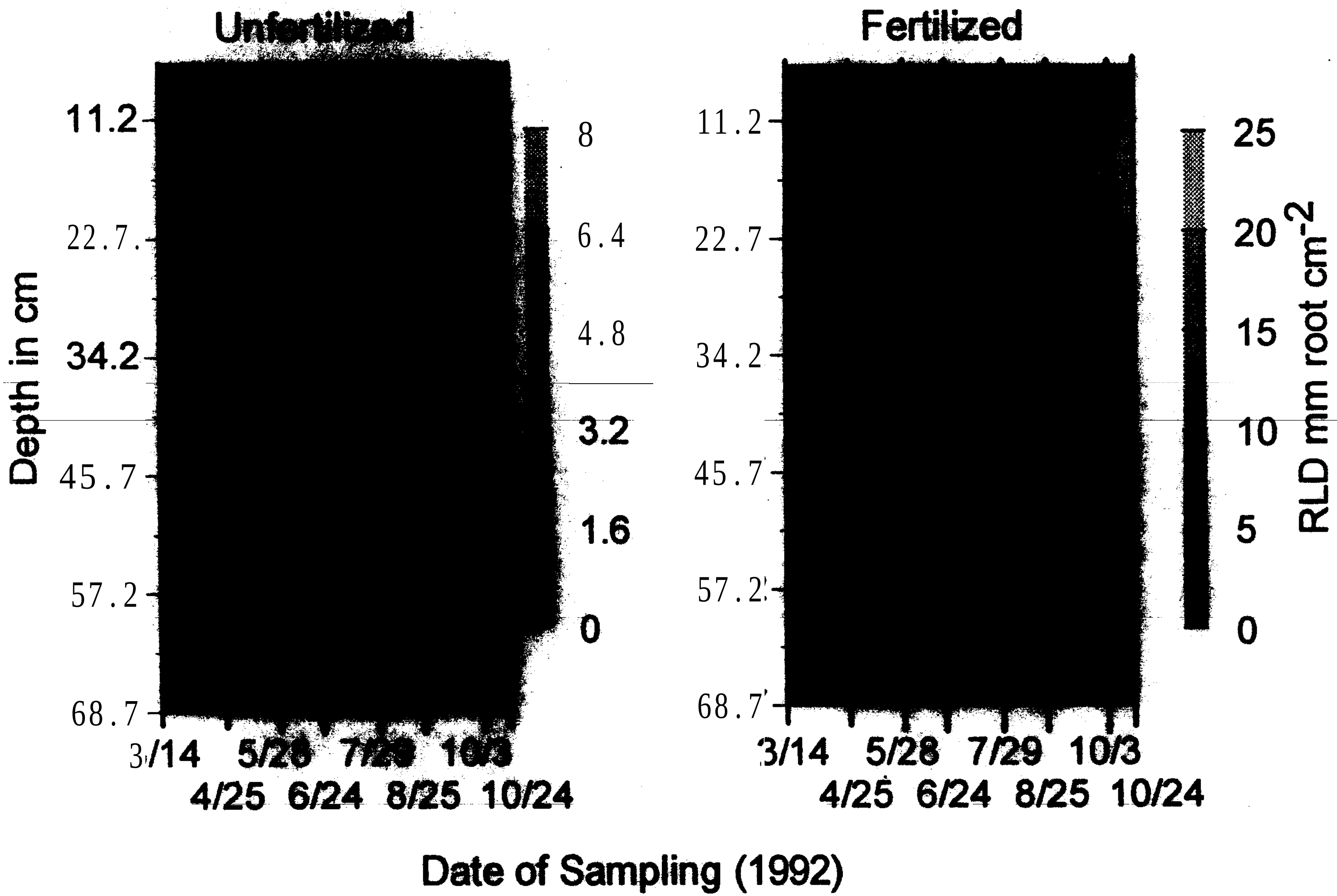
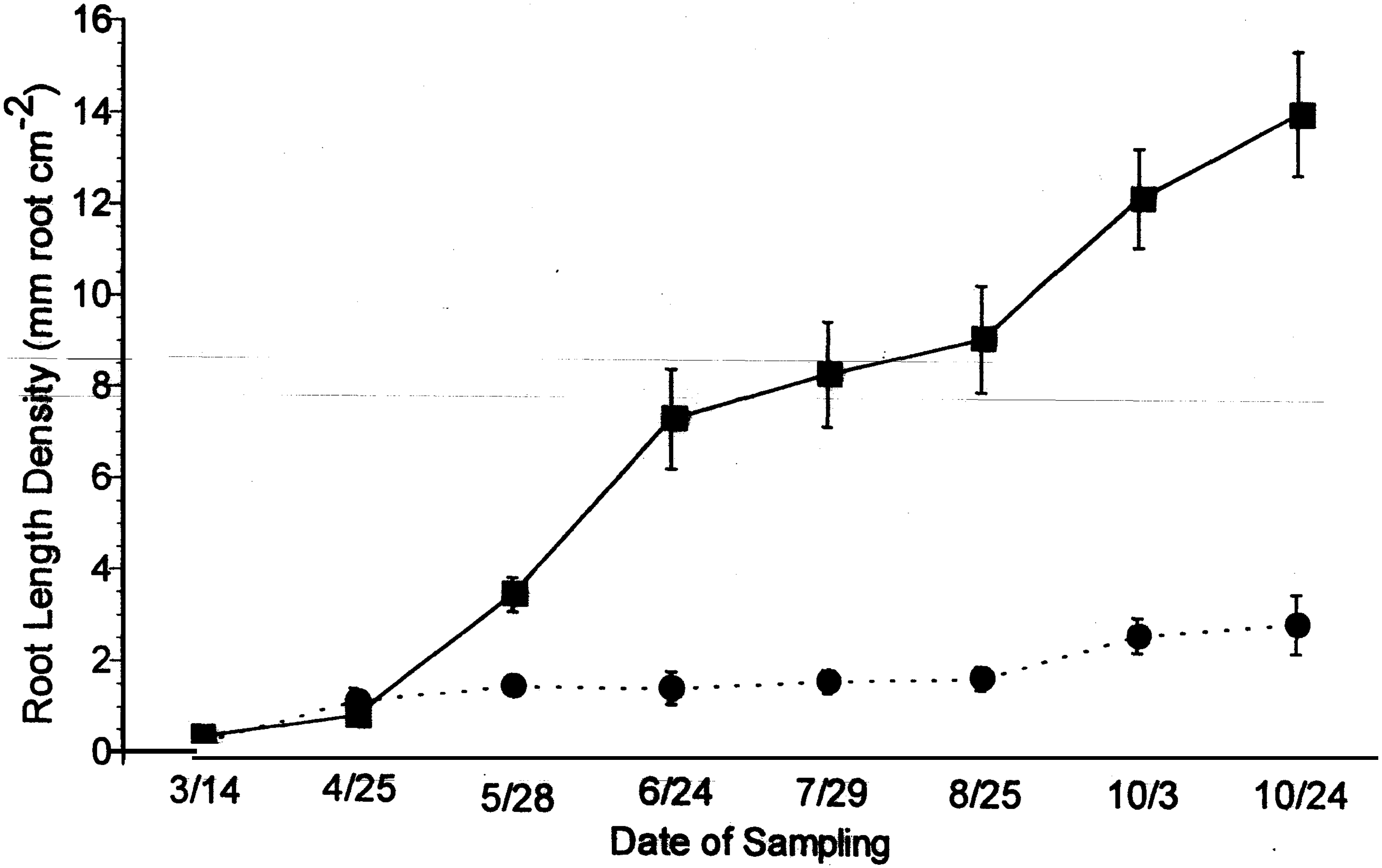

Unfertilized
Fertilized
Depth in cm
11.2
22.7
34.2
45.7
57.2
68.7
8
6.4
4.8
3.2
1.6
0
25
20
15
10
5
0
RLD mm root cm-2
3/14 5/28 7/29 10/3
4/25 6/24 8/25 10/24
Date of Sampling (1992)

Figure 6. Root length density for fertilized plots (■) and unfertilized plots (●) from March 14, 1992 to October 24, 1992. Means and standard errors calculated from mean of plots, with root length density from entire tubes used to calculate plot means.


Root Length Density (mm root cm-2)
16
14
12
10
8
6
4
2
0
3/14
4/25
5/28
6/24
7/29
8/25
10/3
10/24
Date of Sampling

the fertilized treatment (root length density = 14.05 mm cm^{-2}, on 1 0/24/92) than in the unfertilized treatment (root length density = 2.88 mm cm^{-2}, on 10/24/92). The depth*date interaction was somewhat less clear (Figure 7). However, the O-20 cm depth class showed a higher rate of increase with time then the two deeper depth classes. The 20-40 cm and 40-60 cm depth class do not appear to be different from each other. The O-20 cm depth class had the highest root length density.

Turnover

The turnover analysis provided fewer significant interactions than did the root length density analysis. The analysis of variance found one main effect and one interaction to be significant (Table 6). The treatment*depth interaction was significant ($F=2.59, P<0.05$). Turnover tended to decrease with depth in the unfertilized plot while the turnover in the fertilized plot showed no clear change from the beginning to the end of the growing season (Figure 8). The fertilized plots appeared to have a lower turnover than did the unfertilized plots in the O-20 cm depth class (fertilized = 0.020 SE 0.0011, unfertilized = 0.024 SE .0013). There did not appear to be a difference between treatments for the other depth classes. Date was a significant main effect ($F=3.31, P<0.05$). Turnover in March was higher than turnover in either April or May (Figure 91.

Cohort Analysis

Figures 10 and 11 show the cohort root number density by date. The patterns and relative values were similar to root length density for the plots

Figure 7. Root length density (RLD) for 0-20 cm depth (■), 20-40 cm depth (●), and 40-60 cm depth (▲) for both fertilized and unfertilized plots. Means and standarb errors calculated from mean of plots. Note that the scales on the y-axes; are different.

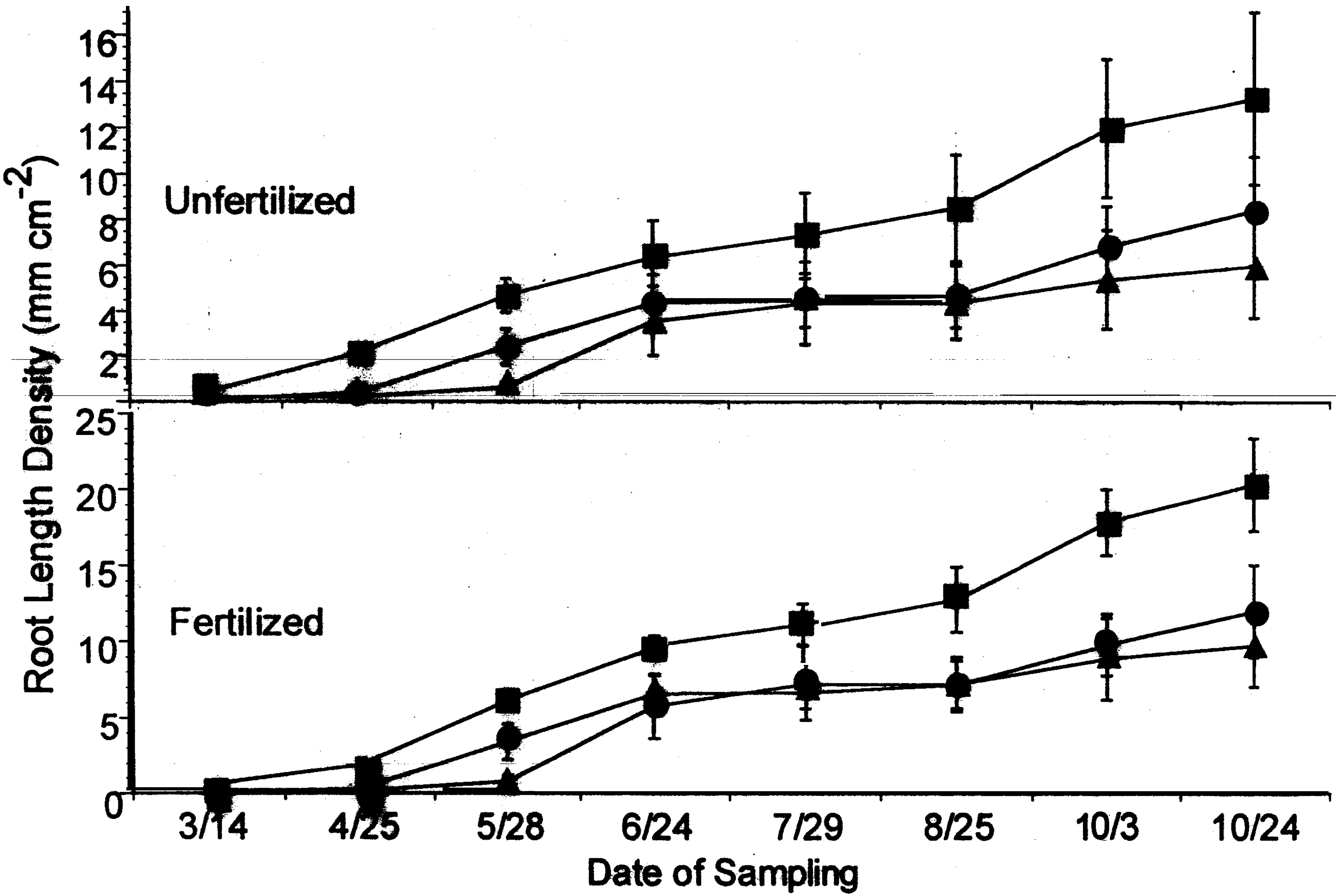

Unfertilized
Fertilized
Root Length Density (mm cm^{-2})
16
14
12
10
8
6
4
2
25
20
15
10
5
0
3/14
4/25
5/28
6/24
7/29
8/25
10/3
10/24
Date of Sampling

Table 6. Nested doubly repeated measures analysis of variance examining the effect of fertilizer on turnover over time and across three depth classes. DF = Degrees of freedom, SS = sums of squares, F Value = calculated F value. SS and Mean square in ten thousandths.

Source of variation	DF	Type IV SS	Mean square	F value	Pr>F
Treat	1	0.061	0.061	0.05	0.8377
Plot(Treat)	3	0.131	0.044	0.03	0.9910
Error	5	6.618	1.323		
Depth	2	2.304	1.152	2.37	0.1438
Depth *Treat"	2	4.613	2.306	4.74	0.0356
Depth*Plot(Treat)	6	1.686	0.281	0.56	0.7410
Error(depth)	10	4.864	0.487		
Date'	3	1.271	0.424	10.76	0.0005
Date *Treat	3	0.120	0.040	1.02	0.4130
Date *Plot(Treat)	9	0.010	0.011	0.27	0.9743
Error(Date)	15	0.069	0.039		
Depth*Date	6	0.170	0.028	1.04	0.4205
Depth*Date*Treat	6	0.295	0.049	1.81	0.1313
Depth*Date*Plot(Treat)	18	0.585	0.033	1.19	0.3261
Error(Depth*Date)	30	0.817	0.027		

. significant at the $p < 0.05$ level.

Figure 8. Turnover for fertilized plots (■) and unfertilized plots (●) for all three depth classes. Means anti standard errors calculated from mean of plots from 6/24/92- 1 0/24/92.

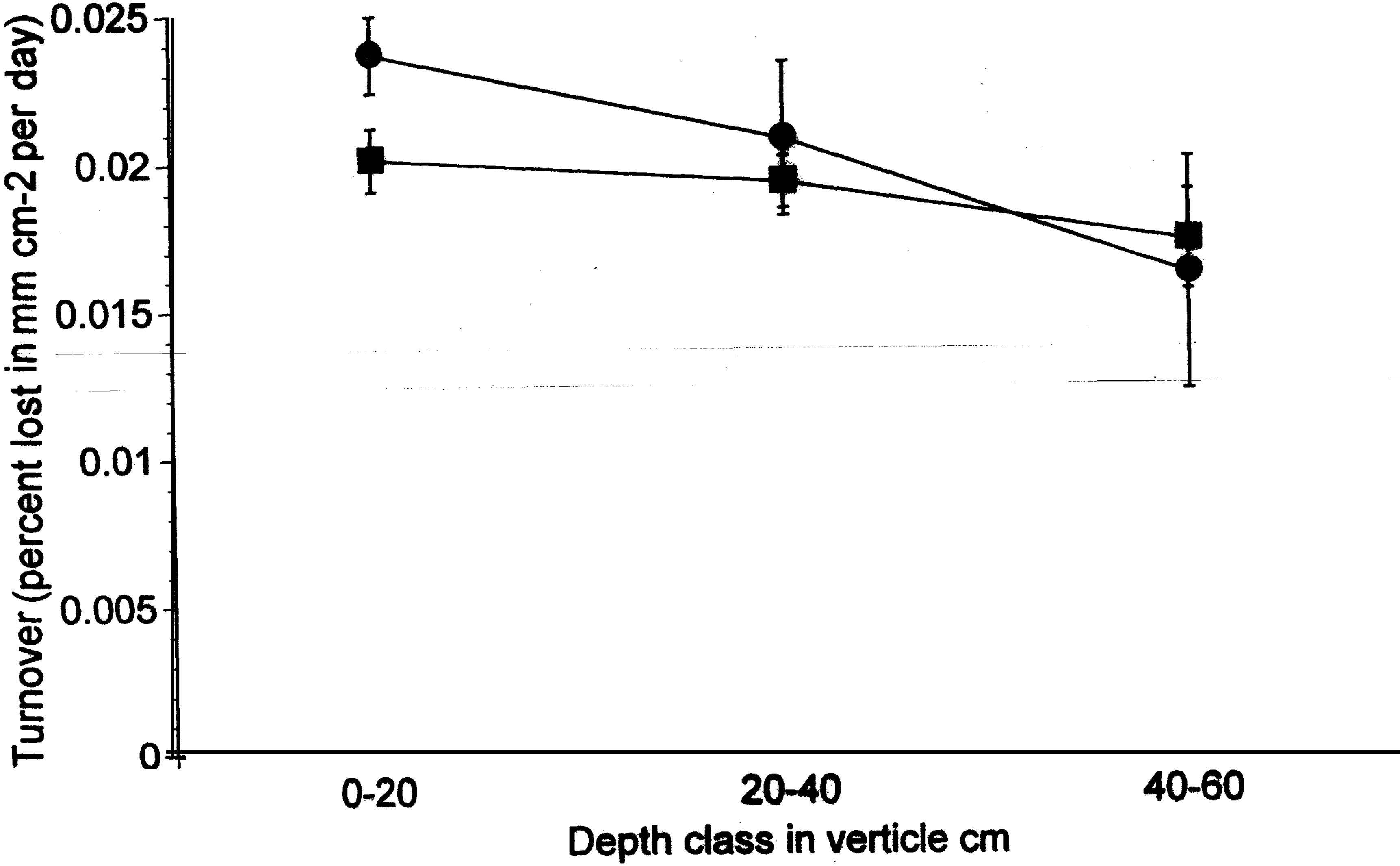

Turnover (percent lost in mm cm-2 per day)
0.025
0.02
0.015
0.01
0.005
0
0-20
20-40
40-60
Depth class in verticle cm

Figure 9. Turnover for date Intervals of fertilized and unfertilized plots. Means and standard errors calculated from mean of plots with root length density from entire tubes used to calculate plot means.

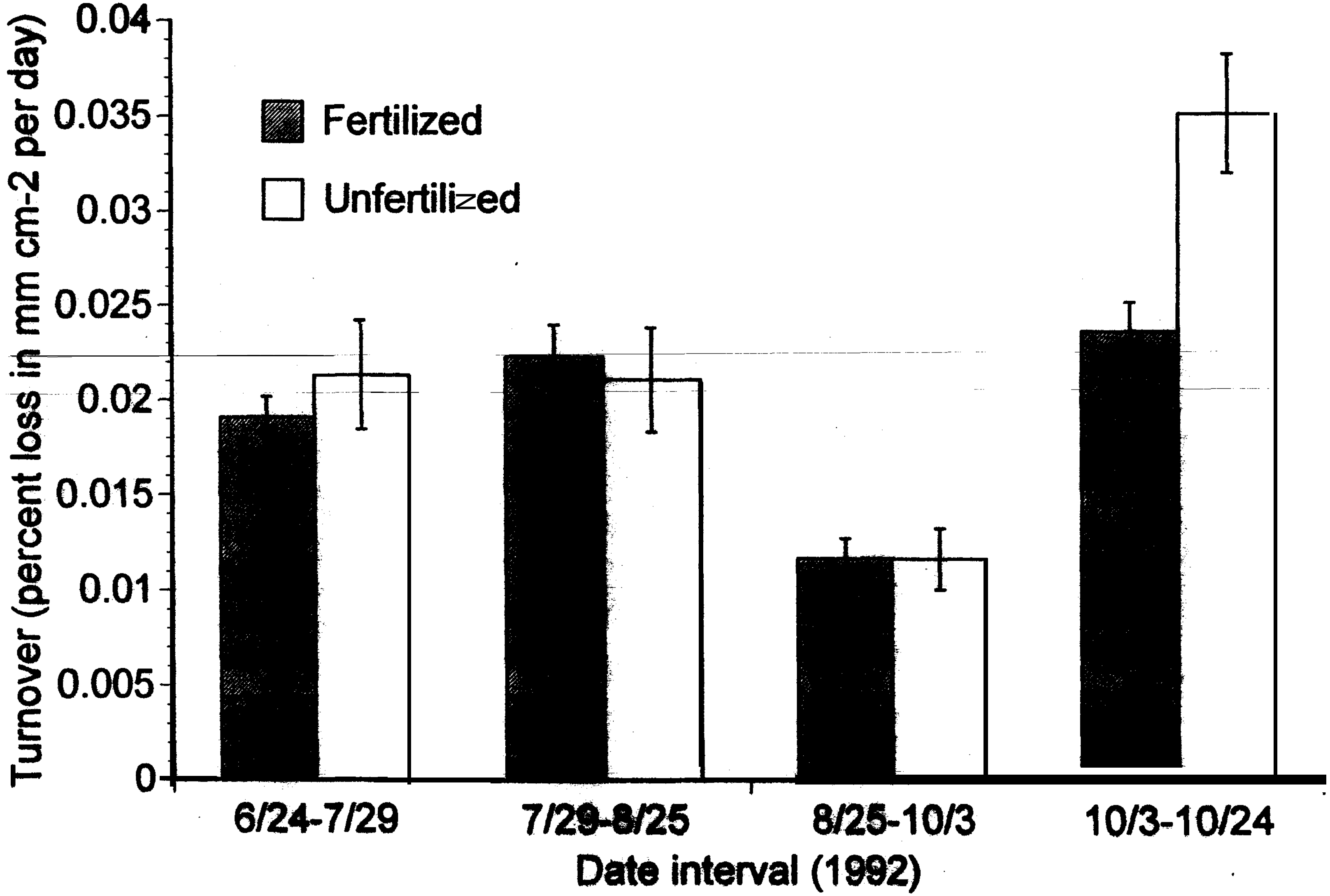
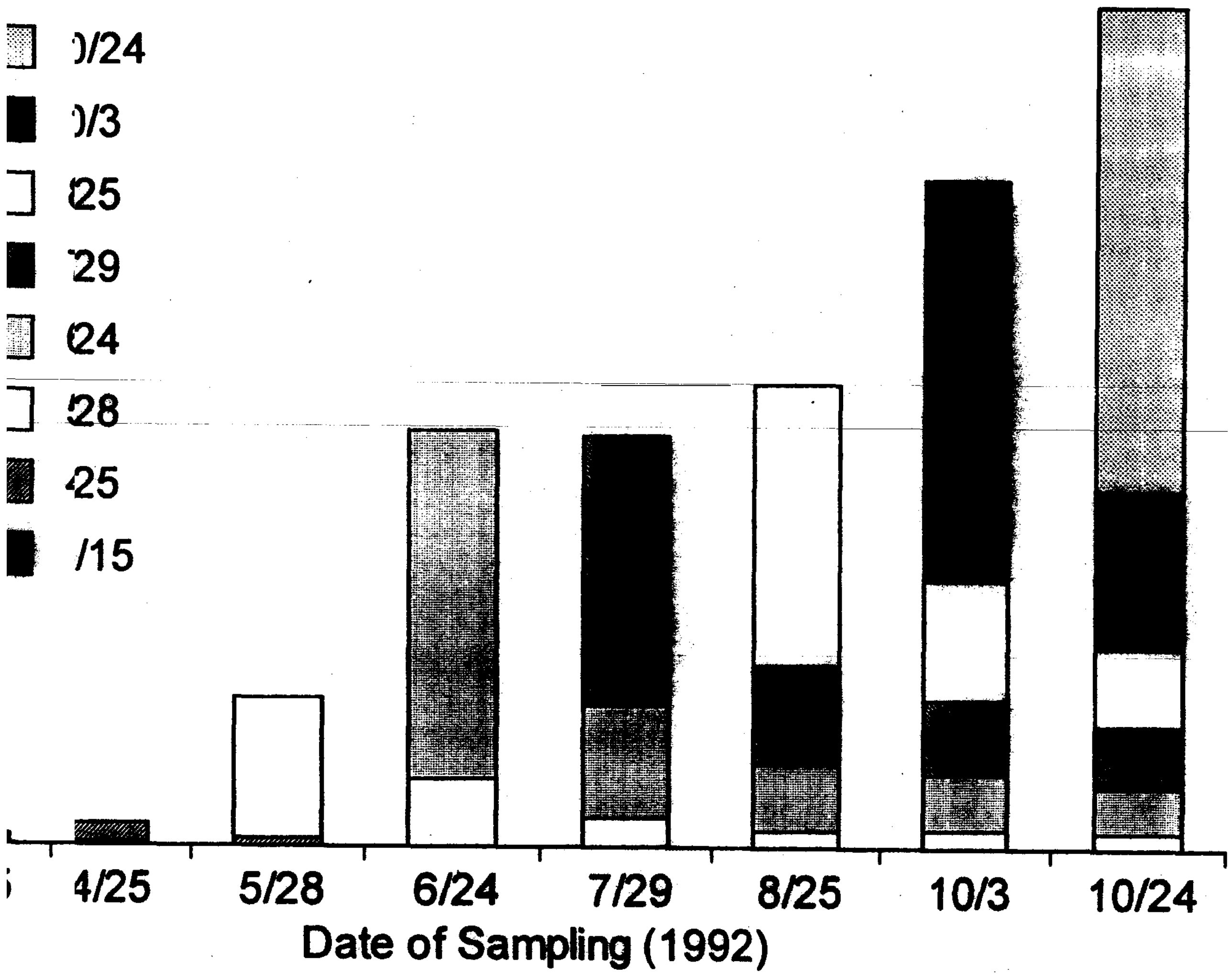
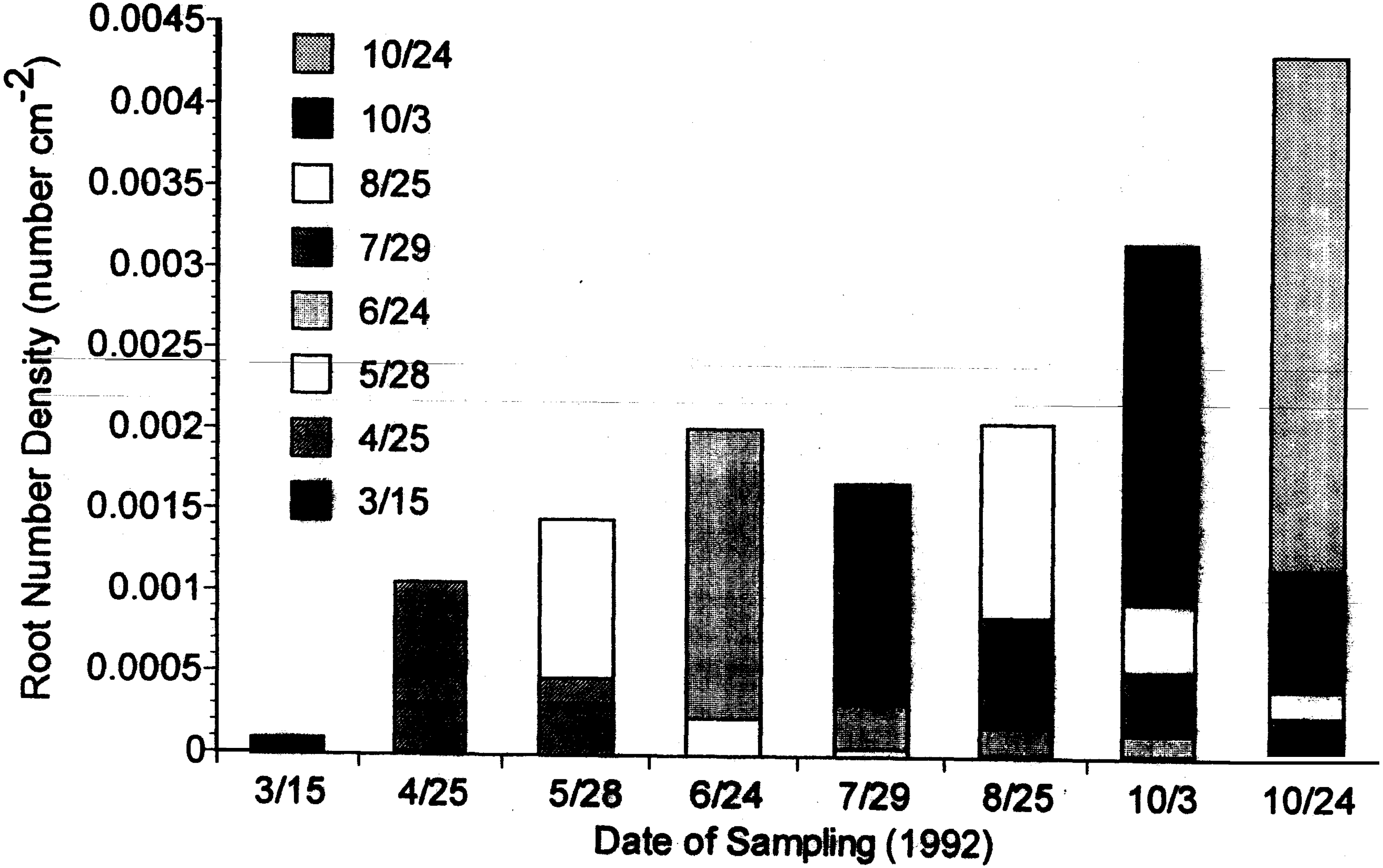

Turnover (percent loss in mm cm-2 per day)
0.04
0.035
0.03
0.025
0.02
0.015
0.01
0.005
0
Fertilized
Unfertilized
6/24-7/29
7/29-8/25
8/25-10/3
10/3-10/24
Date interval (1992)

Figure 10. Cohort data by date for fertilized plots. Each color represents a cohort. Each column represents a sample date.



/24
/3
25
29
24
28
25
/15
4/25
5/28
6/24
7/29
8/25
10/3
10/24
Date of Sampling (1992)

Figure 11. Cohort data by date for unfertilized plots. Each color represents a cohort. Each column represents a sample date.



Root Number Density (number cm-2)
0.0045
0.004
0.0035
0.003
0.0025
0.002
0.0015
0.001
0.0005
0
10/24
10/3
8/25
7/29
6/24
5/28
4/25
3/15
Date of Sampling (1992)

seen in Figure 6. There was an increase in root number density throughout the growing season for both the fertilized and unfertilized plots. Looking at the individual cohorts, one will notice that cohorts have their largest decline in the second observation of the cohort.

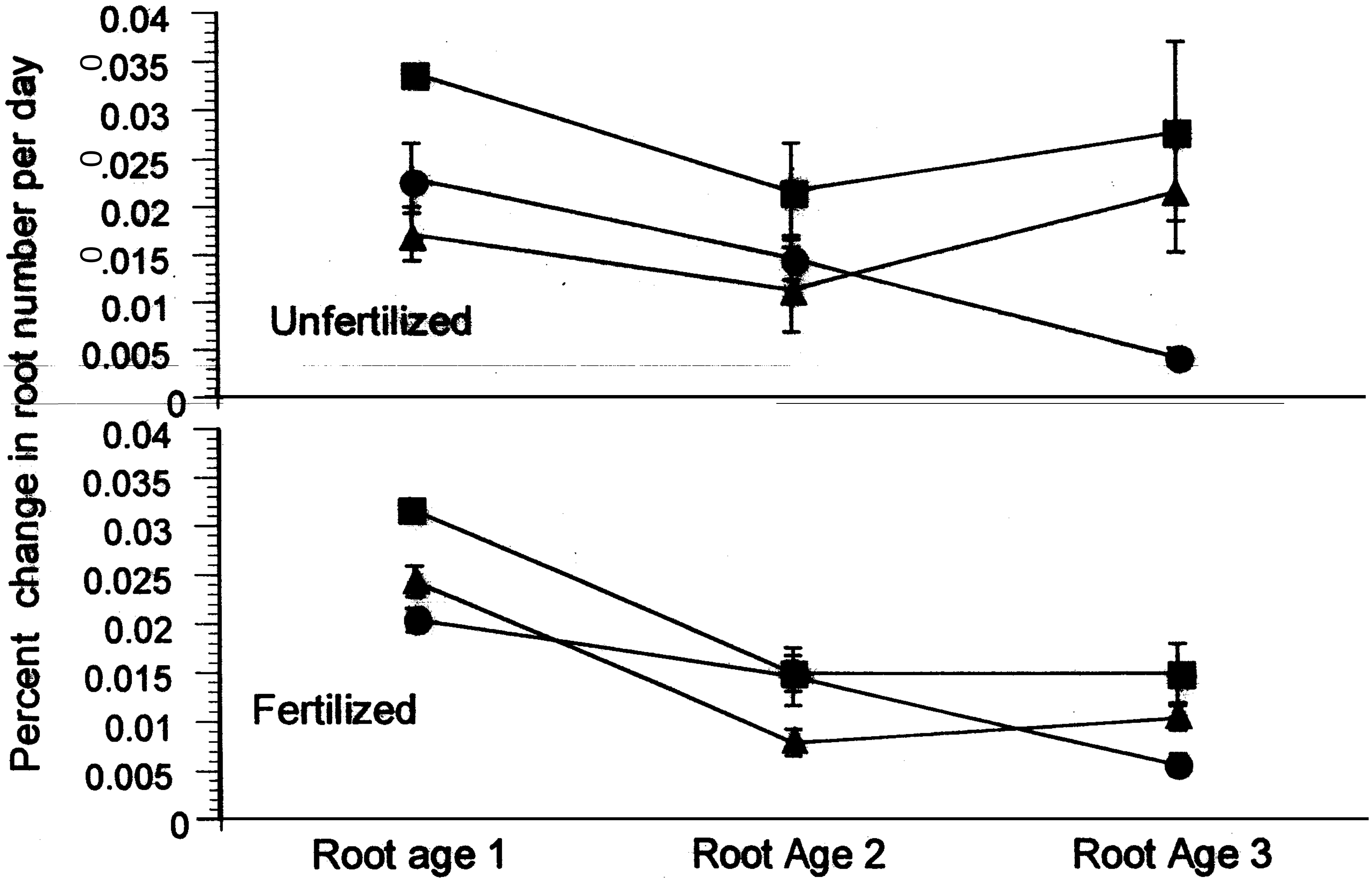
Both the May and June (cohorts had similar patterns with root age (Figure 12). The results for cohort analysis found one interaction to be significant (Table 7), cohort*root age ($F = 1082.21$, $P > 0.0001$) as well as the cohort and root age main effects (cohort $F = 1645.06$, $P > 0.0001$; root age $F = 949.72$, $P > 0.0001$). The first root age was higher than root age categories two and three. Root age's two and three did not appear to be significantly different from each other. Cohorts therefore lost most of their roots between the first and second time they were observed. The cohort*root age interaction was significant because the July cohort evidenced a different pattern from the other two cohorts. The last observation of the last cohort had a lower daily percent change than the second root age category. The other cohorts both showed either a slight increase in root mortality rate or equal root mortality rate in the third root age category.

Discussion

Root Length Density

The nitrogen fertilization only affected the root length density by causing an increase in the fertilized plots. The increase in root length density was expected. Both aboveground and belowground biomass responded similarly in

Figure 12. Cohort analysis percent change per day by root age category and cohort for both fertilized and unfertilized plots. (May cohort(■)), June cohort (●), July cohort (▲)). Means and standard errors calculated from mean of plots with root length density from entire tubes used to calculate plot means.



Percent change in root number per day
0.04
0.035
0.03
0.025
0.02
0.015
0.01
0.005
0
Unfertilized
Fertilized
Root age 1
Root Age 2
Root Age 3

Table 7. Nested crossed repeated measures analysis of variance examining the effect of fertilizer on cohorts across three root age classes. DF = Degrees of freedom, SS = sums of squares, F Value = calculated F value.

Source of variation	DF	Type IV SS	Mean square	F value	Pr>F
Treat	1	0.00002	0.00002	0.03	0.8620
Plot(Treat)	5	0.131	0.0008	1.16	0.3433
Cohort	2	2.306	1.153	1645.06	0.0001
Treat *Cohort	2	0.0002	0.0001		
Error	49	0.034	0.0007		
Root age*	2	1.919	0.960	949.72	0.9001
Root age *Treat	2	0.0008	0.0004	0.40	0.6701
Root age *Plot(treat)	10	0.015	0.001	1.44	0.1755
Root age *cohort*	4	4.373	1.094	1082.21	0.0001
Root age *Treat *Cohort	4	0.001	0.0002	0.28	0.8891
Error(Root age)	98	0.099	0.001		

* significant at the $p < 0.05$ level.

previous studies that used the same fertilization regime (Day, unpublished). There is also a large compendium of evidence that plants respond to fertilization through increased root mass. Titman and Wedin (1991) found an increase in root biomass correlated with increasing nitrogen status for five perennial grass species. In addition, Wilson and Tibnan (1991) found increasing root biomass with nitrogen supply in a study of competition along a nitrogen gradient.

Roots respond more strongly to the higher nitrogen content of the upper portion of the soil. Studies by Drew (Drew and Saker, 1975; Drew, 1975; Drew et al., 1973) found increases in lateral root initiation with N placed within a specific horizon of the soil. Eisenstat and Caldwell (1988) also found similar increases when they added nitrogen at a specified depth. The fact that there was not a significant difference in root density distribution by depth between the control and fertilized plots may reflect Grime and Campbell (1991) and Sharpe and Rykiel's(1991) assertion that resource poor plants, such as those found on Hog Island, may have a less flexible response to nutrient additions.

The dune ecosystem had considerably lower root length density than other systems. Hendrick and Pregitzer (1992*a*) found densities between 1.5-4.5 cm cm^{-2} in a northerdward ood forest. Atkinson and Fogel (1991) also found higher values (4.8 - 18 cm cm^{-2}) along a rhizotron in a northern hardwood forest with thes plants djacent to the rhizotron being *Prunus pumila* and *Pteridium aquilinum*. This fits with Gleason and Tilman's finding that root densities increase in later succesional stages (Gleason and Tilman, 1991).

Turnover

There was a lower turnover rate at the nitrogen rich surface of the fertilized plots than in the less rich unfertilized plots. This finding contrasts with the findings of Aber et al (1985) Who found increased turnover with nitrogen. However, turnover comparisons in Aber et al.'s study were not made between sections of a single soil profile and differences were observed between ecosystems. The plants may be storing nitrogen sources as suggested by Chspin (1980), Grime and Campbell (1991) and Sharpe and Rykiel (1991). Fertilization, therefore, slowed twrnover by changing roots from foraging to storage agents. The overall low turnover is supported by Grime and Campbel's (1991) theory that low' resource high stress plants should have roots with a long functional life.

The changes observed in turnover by date were directly correlated to the number of days between sampli g. I suspect that it is an artifact of the calculation of turnover rather than a true measure of turnover. It may be that there is an inititial turnover that happens at a shorter time period than the sampling interval, as a result the turnover rate may be an underestimate.

Cohort Analysis

There were at least two classes of roots which were observed in the cohort analysis. The initial large turnover followed by two lesser turnovers: for each cohort shows a differential shedding of roots dependent upon the age of the roots. Ephemeral roots w ee observed in this study. This finding is supported by Caldwell (1979). Most of the roots die in the first month. It may be that turnover was low for the last sample period in the last cohort (July Cohort) because it was late in the season. The mean temperature and total rainfall for October (the period for turnover in the last cohort) was considerably lower than in the previous months (October 14.7°C/44.6 mm; Sept. 21.9°C/162.8 mm). Hendrik and Pregitzer (1993) found that cohort turnover declined over the winter months. Garwood (1964) also found that roots initiated late in the season were longer lived in a variety of agricultural systems. Because these plants are low resource perennial plants, they may have been maintaining their roots for the next spring. The roots will, therefore, act 8s plant reserves and maintain resources for the plant for future us8 (Grime and Campbell 1991, Sharpe et al. 1991).

Conclusion

Patches of nitrogen fertilizer appeared to ellicit a whole plant response rather than a singular strong response by roots within the fertilized zone of the soil. The only response to the difference in N fertilization with depth was in turnover. Grime and Campbell (1991), Sharpe and Rykiel (1991) and Grime (1977) felt that resource poor (plants, such as those found on Hog Island, may have this less flexible response to nutrient additions. The overall low turnover rate, the decreased turnover rate with fertilization and the decreased turnover in the last cohort imply that roots tend to be conserved in this system. This implies that the metabolic maintenance and resource storage of the roots may be more important than foraging by roots in resource poor plants.

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Protocol for digitizing plant roots with Roots software

The following procedu 8 will allow the user to digitize roots through repeated digitizations of the s3me window. To assist in this process a method has been developed for file namingIt is important to develop a similar method for your files. This aids in file manipulation and recall for analysis. Through the careful use of' the ? and • wildcards all files can be found for each month, tube, frame.

There are three types of fiies generated by ROOTS.

- .f.AB fife containing length, width, root_id, etc. (dbase)
- *.arc file containing outlines, skeletons, or points (dbase)
- *.id file which gives max height and width of frame (ascii)

There are three major record pointers contained within the • .fab fife. They are Root_id, Seg_id, and Arc_id. The Root-id is a sequential number generated by the roots software. This uniquely identifies each observation of each root. Arc_id is also generated by the software. This tells the software which record starts or ends the outline or skeleton of the root in the *.arc file. The seg_id pointer is the same as the root-id unless the user changes this number after dfgitfzatfon. My method uses the seg_id to. record whether a root was observed in prior observations and which root was observed in the prior sampling.

It is important that the user have access to a database program which uses dbase files as its native format. These include, dbase III plus and dBase IV, and Microsoft foxpro. There are probably more that exist but these are the only ones I have tested with this data. ROOTS has an extremely limited ability to edit the data once the root has been digitized. Therefore corrections will need to be made using the browse mode of one of these other software packages after completing the digitization. In addition, some programs have been developed which may assist in the compilation and manipulation of the ROOTS data. The programs have some documentation but some modifications will probably need to be made to tailor the programs to your specific system and needs. Feel free to cannibalize, augment, or otherwise after the code in these provided programs. As stated in the prologue to the programs, these programs are free and there is no warrantee or guarantee associated with programs. I have tried to make them conform to the rules of dBase III plus, the lowest common code denominator.

After installation of the software some adjustments need to be made to optimize the use of ROOTS. ROOTS places all of the root data for each frame in its own file. Although tidy, this provides theenduser with a speed problem.

After about 99 files the hard drive really slows down on file access. Because three files are generated for each frame digitized, the number of files in your data directory increases quickly. Therefore a protocol was developed in which 8 work directory is used for the active tubes relevant files. If you do not experience 8 slow down in processing speed, you can of course ignore this and related portions of the protocol.

Method for accessing new data directories and setting up work file

The roots software assumes that you are going to use the DATA subdirectory of the CMAP subdirectory. Therefore if you would like to change the location of the active directory in which roots starts you must alter ?????.ini. Use edit or edlin to change the default directory to where you would like it to start. I have set my ?????.ini file to have c:\cmap\work 8s my default directory. I keep stored data in other subdirectories of the cmap subdirectory. After I set up roots I copy the relevant files from the appropriate subdirectory into the work directory. After I finish each tube, I copy the newly created files to the appropriate storage subdirectory (for instance c:\cmap\nasa for my nase study). I then delete all of the *.arc, ● .id, and ● .l8b files end copy in the new files for the next tube. Be careful not *to* delete the scale file which holds info on the scale of the frame.

movement

setting up scale

listing for each option on roots screen (and purpose)

Protocol for ROOTS software

1) start roots
2) move to scale
3) type y (to start scale selection)
4) chose 3 (previous setup)
5) enter name of scale file (it must be in your current dir)
6) click on the right mouse button (to accept setup)
7) type y (the scale is acceptable)
8) type 6 enter (to exit scale setup)
9) move to date
10) enter date of video sampling
11) move up to site, plot, and tube and set for first sample
12) move to extent
13) type O (to select options)
14) tab down to edit
15) type y (to allow editing of record)
16) press esc (to leave wirow)
17) move to order
18) type y
19) chose order fo movement (2,7,3,13,14,1)
20) move to capture
21) press y
22) start video,
23) when you have the correct image press spacebar
24) enter the frame number
25) enter the tube and frame number in your paper lab book
26) if there is a prior image press y
    26a) press y (use scale)
    26b) accept the rest of the defaults (press return)
27) enter the filename of the frame to be digitized

use the format
date d; site ss; frame fff; tube tt
filename dssffftt
example ahg03713

date	a	3/14/92
site	hg	(hog island)
frame	37	37th frame from bottom
tube	13	tub8 13 in plot 3, a fertilized plot

28) type y (make sure you are in extent)

- 28a) *move mouse to tip or base* of root and click left mouse button
- 28b) click up length of root with left mouse button
- 28c) at last point click the right mouse button
- 28d) if you accept the length press the spacebar otherwise press esc and start this root over
- 28e) find an area of representative width on the root
- 28f) click right mouse button
- 28g) follow width with right clicks
- 28h) end width measurement with left 'mouse button click
- 28i) press spacebar to keep measurement or press esc to repeat width measurement
- 28j) position crosshair where you would like root-id and click left mouse button
- 28k) enter a single character code for root condition or other parameter and press return
- 28l) if you need to check other records within this file do so now, one can also make corrections.
- 28m) if the root digitized is the same as from a prior digitizing episode, enter a negative sign and that prior roots root_id (ie -7 for a root with a root_id of 7 in the previous month) and press return.
- 28n) press esc when finished editing

28) press y (to blank and reset screen)

30) press y (to start capture)

31) to end procedure press esc and then y to exit.

Capture:

This option allows the user to capture 8 video image from the video input. When the user enters Y, the video image appears on the second (video) monitor. While the video is rwnning the user can adjust the images contrast and brightness levels through the use of the arrow keys. After the image is captured adjustments cannot be made to the quality of the image. To achieve the clearest image it is best to capture the image while the video is still. if there is movement, the captured image will vibrate end the image will probably be stretched in the direction of the camera movement. To freeze an image, press the space bar. When the space bar is pressed the cursor on the ROOTS screen moves to the next option.

Display:

this option 8iiows the user to display on the second (video) monitor 8 stored video frame.

Save:

This option allows the user to save a frame on the' harddrive for use with display. This option takes wp lots of hard drive space and is not recommended for routine use.

Blank:

This option allows the user to clear the second (video) monitor and reset the poeition conditions which may have been altered with the zoom feature. it is suggested that the user use the blank option prior to digitizing each frame to ensure that the system has been reset. A possible result of not blanking prior to digitizing is improper length, width, and perimeter measurements and improper placement of outlines, skeletons, or points.

Frame, Tube, Plot, Site, Date:'

These options hold information on the current frame related to placement and date of trampling. At start up, FRAME, TUBE, PLOT, and SITE, will be 1. The date may be 6\24\89. To change the values, simply move to the option associated space and enter the new information. This is the data which is placed in the *.lab file.

File:

This option provides the name of the file to record the information obtained through digitization. (See naming conventions in the introduction)

Draw:

This option allows the user to produce a previously digitlzed frame on the video monitor.

Attributes:

This option allows the user to change the colors and sizes of the text and lines used for digitizing roots.

DOS:

This option allows the user to enter DOS. Most of the memory is used for the roots program. Therefore only limited activities can be performed here such as copying and renaming files.

Areas:

This option allows the user to obtain and record length, width, and perimeter information from a video frame. Unless one requires the outline rather than the skeleton or requires an area calculation which cannot be obtained from the length and width, it is suggested that extent be used instead of area. Extent takes less time per root.

Length:

This option allows the user to obtain length measurements from the video image. No width calculation will be made.

Extent:

This option allows the user to obtain both length and width measurements from the captured video image.

Points:

This option allows the user to obtain the coordinates of polnts of interest from the captured video images. I have seen this option used to track insect behavior in a rhizotron.

Mapsetup:

This option allows the user to set up the scale to be used for the AREA, LENGTH, EXTENT, and POINTS calculations. One must have a scale set in order to obtain measurements in units of measure rather than in pixels.

Sequence:

This option allows the user to set up the order in which the options are accessed. The user can then move from frame to draw to file to extent to blank to capture. This provides a much more efficient method of movement than simply tabbing through the options.

```
** program curve.prg - this program determines peak position and height.
* The program assumes that there is a unimodal distribution. The program
* moves down the profile and sums each succesive three sections. The
* position for the peak is the center section.
* written by Everett Weber written 9/13/94 | revised 9/13/94
close all
sele 2
use peak

sele 1
use (section
index on dtoc(date)+' '+tube+str(section) to tube

do while .not. eof()
          m_date=Date
          m_tube=tube
          peak_RLD = 0
          peak_sec = 0
         Do while tube=m_tube
          if section>1
            skip -2
          endif
           x=1
           sec_RLD = 0
          do while x < 4
           if x = 2
                    m_sec = section
           endif
             sec_RLD = RLD + sec_RLD
           if x=3
              if sec_RLD > peak_RLD
                 Peak_RLD = sec_RLD
                 peak_sec = m_sec
              endif
           endif
           x=x+1
           skip
          enddo
          sele 2
          append blank
          replace tube with m_tube
          replace RLD with peak_RLD
          replace depth with peak_sec
```

```
        replace date with m_date
        sele 1
       enddo
enddo
```

```
** Cohort.prg - this program is designed to provide information on
*       for the cohort analysis. It counts roots of each cohort
*                       within each tube. A filter has been set to eliminate
*                       unneededcohorts and unneeded dates of needed cohorts.
*                   A new database (cohort) is used to store the current
*                       countandthencxt months count for each tube as well as
*                       the proportional difference between months. Date in the
*                       new database is actually the sample number form the first
*                       instance of siting of the cohort. There are therefore only
*                       4 dates (1-4) in the dataset. See methods section of thesis
*                       for more info on these calculations or read the program.
* Program written by Everett Weber on 10/27/94 | Last revision 10/27/94
Set talk off
set safety off
clear
Sele 3
use dates
index on month+day+year to date

Sele 2
use cohorfl
zap

@1,1 say 'indexing main database and setting up filters and relations'
sele 1
use hog
set relation to month+day+year into c
  * eliminate unneded portions of database
set filter to (val(init)=3 and val(c->datecode)<7) or (val(init)=4 and val(c->datecode)<8) or
val(init)=5

* count roots in each date,tube,init group
index on c->datecode+tube+init to tube
go top

clear  &&clears screan
@ 1,1 say 'inputing data into cohort.dbf'
 do while .not. eof()
      m_init=init
      m_tube=tube
      m_date=c->datecode
      count=0
      @ 2,2 say 'date = '+m_date
```

```
        @ 3,2 say 'init = '+m_init
        @ 4,2 say 'tube = '+m_tube
   do while c->datecode=m_date and tube=m_tube and init=m_init
        count=count+1
        skip
   enddo
    sele 2
    append blank
    replace tube with m_tube
    replace init with val(m_init)
     n_date = 1+(val(m_date)-val(m_init))
    replace date with n_date
    replace n with count
    sele 1
  enddo

sele 1
use

Clear

sele 2
index on str(init,3,0)+tube+str(date,3,0) to tube
go top
  @ 1,1 say 'calculating cohort turnover'
 do while .not. eof()
        m_rec=recno()
        m_tube=tube
        m_init=init
        m_date=date+1
        @ 2,2 say 'date = '+str(m_date,3,0)
        @ 3,2 say 'init = '+str(m_init,3,0)
        @ 4,2 say 'tube = '+m_tube

        skip
  if m_tube=tube and m_init=init and m_date=date
              m_n2=n
        else
          m_n2=0
  endif
        skip-1
        replace n2 with m_n2
        m_change=((n-n2)/n)   && need to calculate days between sampling
```

```
    replace change with m-change
    skip
    if date4 and .not. eof()
  skip
  endif
enddo
```

```
** combine.prg — this program combine the data from all chosen
*  roots databases and places them into DBF hog1

set safety off
set talk off
sele 1
 use f_data
sele 2
 use hoge
 zap
 go bott
 sele 1
do while .not. eof()
   file=fil_name
   sele 3
   use File+'.lab'
     go bott
     rnumb=recno()-1
   sele 2
     append from file+'.lab'
     go bott
     skip -rnumb
    do while .not. eof()
     replace source with file
     skip
    enddo
   sele 1
   skip
enddo
close all
sele 1
use hoge
set safety on
set talk on
→
```

```
** program cluster.prg - this program sums all root within a specified
* section of the tube. Output from this file can then be used in other
* programs to find the RLD peak or other analyses. The program uses the
* file output.dbf as input and creates the file section.dbf.
* created 9/20/94 by Everett Weber; revised 9/20/94

close all

sele 2
use cluster
zap

sele 1
use section
set filter to section<13 && eliminates sections greater than 12
index on dtoc(date)+tube+str(section) to tube
go top

m_sec=4
m_tube=tube
m_date=date

do while .not. eof()
        M_RLD=0
        do while tube=m_tube and m_date=date and m_sec>=section
         M_RLD=M_RLD+RLD
         skip
        enddo
  sele 2
        append blank
        replace tube with m_tube
        replace date with m_date
        replace section with m_sec/4
        replace RLD with M_RLD
  sele 1
        m_tube=tube
        m_date=date
              m_sec=m_sec+4
       if m_sec>12 or date<>m_date or tube<>m_tube
         m_sec=4
       endif
enddo
close all
```

```
*program labdir.prg - this program creates a list of all dbf files and creates a single file which
*                     combines all of the files togather. One can substitute *.lab for *.dbf to
*                     combine all of the *.lab files into a single database.
set talk off
sele 1
!del lab.dir
!dir *.dbf>lab.dir

use f_data
zap
append from lab.dir sdf
   go top
   dele
   skip
   dele
   skip
   dele
   skip
   dele
   skip
   dele
   go bottom
   dele
 pack
```

```
*** inception.prg -- this program determines the date at which a
* root initially entered the viewing area of a minirhizotron frame.
* This program requires that the roots are tracked in the seg_id
* field as described in the acompanying document.
* created 2/27/94 | revised 7/25/94
** do labid
** do combine (combined file has added field init)
set talk off
set safety off
sele 3
use dates
index On month+day+year to date
sele 2
use error
zap
sele 1
use hog7
flag=.f.
set relation to month+day+year into dates
index on tube+frame+c->datecode to tube
do while .not. eof()
    if seg_id>0
      replace init with c->datecode
    else
      m_frame=frame
      m_tube=tube
      m_root=root_id
      m_seg=seg_id
      d_code=c->datecode
      m_month=month
      m_day=day
      m_year=year
      oldrec=recno()
*       find record just prior to current date+tube+frame combination
     do while (val(c->datecode)=val(d_code) and frame=m_frame and tube=m_tube and .not.
bof())
        skip -1
     enddo
      * find record just prior to privious datecode's date+tube+frame combination
     do while (val(c->datecode)=val(d_code)-1) and frame=m_frame and tube=m_tube and .not.
bof())
        skip -1
     enddo
```

```
* skip to place pointer at start of set
if .not. bof()
  skip
endif
* find specific record from previous sampling
do while root_id<>(m_seg*-1) and val(c->datecode)=val(d_code)-1
skip
enddo
* place init code into memory variable
if root_id=(m_seg*-1) and val(c->datecode)=val(d_code)-1 and frame=m_frame and
tube=m_tube
  m_init=init
else
 * if couldn't find root-id, use current init code (ie. month of sampling)
 goto oldrec
 m_init=c->datecode
 sele 2
 * place record in error file to let user know where problem occured
 * may be an error with the database.
  append blank
  replace error with 'root-id not found'
  replace root-id with m_root
  replace month with m-month
  replace day with m_day
  replace year with m_year
  replace tube with m_tube
  replace frame with m_frame
 sele 1
 flag=.t.
endif
goto oldrec
* placing initiation datecode into record from mem variable
replace init with m_init
endif
skip
enddo
if flag
 ? "there has been an error during execution of the program..."
 ? "please check the DBF file ERROR where these errors"
 ? "have been recorded"
endif
sele 1
close all
```

```
** program sectsum.prg - this program sums all root within a specified
* section of the tube. Output fro this file can then be used in other
* programs to find the RLD peak or other analyses. The program uses the
* file output.dbf as input and creates the file section.dbf.
* created 9/13/94 by Everett Weber; revised 9/13/94

close all

sele 3
use sectcoh
zap

sele 2
use frame
index on tube+frame to tube

sele 1
use output
set relation to tube+frame into b
index on month+day+year+tube+str(b->section)+init to tube2
* set index to tube2
go top

m_init=init
m_depth=b->section
m_tube=tube
m_date=month+'/'+day+'/'+year
do while .not. eof()
        M_RLD=0
        do while tube=m_tube and m_date=month+'/'+day+'/'+year and m_depth=b->section and
init=m_init
          M_RLD=M_RLD+length
          skip
        enddo
  sele 3
        append blank
        replace init with m_init
        replace tube with m_tube
        replace date with ctod(m_date)
        replace section with m-depth
        replace RLD with M_RLD
  sele 1
   m_init=init
```

```
        m_depth=b->section
        m_tube=tube
        m_date=month+'/'+day+'/'+year
enddo
close all
```

```
** program peak.prg - this program determines peak position and height.
* The program assumes that there is a unimodal distribution. The program
* moves down the profile and sums each succesive three sections. The
* position for the peak is the center section,
* written by Everett Weber written 9/13/94 | revised 9/14/94
close all
clear
bot_sec=13
@ 5,2 say 'please enter the bottom section number'
@ 4,2 get bot_sec picture '999'
read

cl ear
close all

sele 2
use peak
zap

sele 1
use section
index on dtoc(date)+' '+tube+str(section) to tube

do while .not. eof()
   @ 7,5 say 'current tube number = '+tube
   @ 8,5 say '             date = '+dtoc(date)
      m_date=Date
      m_tube=tube
      peak_RLD = 0
      peak_sec = 0
     Do while tube=m_tube
      test=.f.
      test2=.t.
      if section>1
        skip -2
      endif
       x=1
       sec_RLD = 0
      do while x < 4
       if x=1 and rld=0
         test=.t.
       endif
       if x = 2
```

```
            m_sec = section
            if rld=0 and test && testing to find out if first two sets are zero
             test2=.f.
            endif
      endif
        sec_RLD = RLD + sec_RLD
      if x=3
        if sec_RLD > peak_RLD
          if (section>bot_sec and test2) or test2
           Peak_RLD = sec_RLD
           peak_sec = m_sec
          endii
        endif
      endif
      x=x+1
      skip
     enddo
    enddo
     sele 2
     append blank
     replace tube with m_tube
     replace RLD with peak_RLD
     replace depth with peak_sec
     replace date with m_date
     sele 1
enddo
```

```
* out.prg -- A program to create an output file to be used by
*            the updat .prg file. the file output contains
*            the format from roots with all dates tube and
*            frames to be analysed. the second work area
*            contains the file listing frame and tube data
*            the third work area contains the dates sampled
*            and the first work area contains the output.
*            The output of this program is used in updat.prg
*    created by Everett Weber created on 07/12/93 | updated 03/16/94
*    of Old Dominion University there is no copywrite for this
*    program and this program should be distributed without
*    any fee or charge. There is also no warentee garontee on this software.
set talk off
sele 1
use output
zap
sele 2
use frame
index on tube+str(section,2,0) to tube
go top
sele 3
use dates
go top
do while .not. eof()
   m_month=month
   m_day=day
   m_year=year
   m_init=datecode
? 'month ='+month
   sele 2
  do while .not. eof()
   m_tube=tube
   m_frame=frame
   m_section=section
   sele 1
   append blank
   replace tube with m_tube
   replace frame with m_frame
   replace arc_id with m-section
   replace month with m-month
   replace day with m_day
   replace year with m_year
   replace init with m_init
```

```
    replace length with 0.000
    replace width with 0.000
   sele 2
    skip
  enddo
    go top
  sele 3
  skip
enddo
sele 2
close all
```

➡

```
• * turnover.prg -- this program calculates turnover on a per cohort
* basis. The program assumes that if a root disapears that the
* root is dead. The mathmatical formula for turnover is
* the same as hendrick and pregitzers mortality rate described in
* thier hardwood forests paper.
*   length mortality/origonal length/time=mortality rate (turnover)
* time is in days for this experiment
* program written by Everett Weber on 10/07/94 | revised 10/15/94
* invaluable assistance debugging was provided by Sharon Haines
set safety off
settalkoff
clear

sele 3
use dates
index on month+day+year to date

sele 2
use turnover
zap

sele 1
use hog

clear

@ 5,5 say 'indexing by date,tube'
index on month+day+year+tube to tube2 unique

@ 5,5 say 'indexing by date, tube, frame'
index on month+day+year+tube+frame to tube3 unique

set unique off
@ 5,5 say 'indexing by date, tube, frame not unique'
index on month+day+year+tube+frame to tube4

set relation to month+day+year into c
go top
clear
do while val(c->datecode)<8
* record data from root at time n

        m_segid=(root_id*-1)
```

```
date=val(c->datecode)
date_store=ctod(month+'/'+day+'/'+year)
m_tube=tube
m_frame=frame

@ 5,5 say ' date = '+dtoc(date_store)
@ 6,5 say' tube = '+tube
@ 7,5 say ' frame = '+frame
@ 8,5 say 'Seg_id = '+str(m_segid,5)
        oldrec=recno()
        n1 length=length
        m_segid=(root_id*-1)
  set index to tube;!
  goto oldrec

do while (.not. eof()) and ((val(c->datecode)<(date+1)) or
(((val(c->datecode)=(date+1)) and (tube<>m_tube))))
  skip
enddo
  if eof()
    skip-l
  endif
  m_recno=recno()
  set index to tube3
  got0        m_recno
do while (.not. bof()) and val(c->datecode)=(date+1) and (tube=m_tube)
  skip-l
enddo
  skip

do while (.not. eof()) and frame<m_frame and val(c->datecode)=(date+1) and
tube=m_tube
      skip
enddo
      if eof()
        skip -1
      endif
  m_recno=recno()
  set index to tube4
  got0 m_recno
do while val(c->datecode)=(date+1) and tube=m_tube and frame=m_frame and .not.
bof()
      skip -1
```

```
          enddo
           skip

          do while seg_id<>m_segid and val(c->datecode)=(date+1) and tube=m_tube and
frame=m_frame and .not. eof()
                skip
          enddo

          if (date+1)<>val(c->datecode) or tube<>m_tube or m_frame<>frame seg_id<>m_segid
or eof()
           turn=1
          else
            mortality=n 1 length-length
           if mortality<0
             turn=0
            else
             turn=mortality/n 1 length
           endif
          endif
          goto oldrec
          skip
 sele2
  append blank
  replace tube with m_tube
  replace frame with m_frame
  replace date with date_store
      replace turnover with turn
      replace root_id with (m_segid*-1)
 sele 1
enddo
```

```
** program trantum.prg - this program transposes minirhizotron data from a
* linear file to a file with each field representing a date depth combination.
* The first part of the program determines the scope and names
* of the fields (first part of the name is the section, the second part of
* the name is the date code). The second portion of the program places the
* data into the created database from the section database created by
* sectsum.prg.
* Written by Everett Weber on 9/14/94 | last revision 9/14/94

close all
clear

sele 2
use dates
index on ctod(month+T+day+T+year) to tube2

sele 3
 use structur
 zap
 append blank
  replace field_name with 'tube'
  replace field-type with 'C'
  replace field_len with 3
  replace field_dec with 0

* note fourth database will be opened called ·anspose
*   this database will contain the transposed data
sele 1
 use clusturn
  set relation to date into b
  index on str(section)+b->datecode to tube unique
  go top
 clear
@ 2,2 say 'creating transpose'
 do while not. eof()
        m_name='dp'+alltrim(str(section))+'_dt'+alltrim(b->datecode)
        @ 3,2 say 'name = '+m_name
  sele 3
    append blank
    replace field-name with m_name
    replace field_type with 'c'
    replace field_len with 10
*   replace field_dec with 3
```

```
  sele 1
        skip
 enddo

sele 5
create tranturn from structur
use tranturn

*   end of file creation section                        **
** ------------------------------------------------------ **
** ------------------------------------------------------ **
*  section which transposes data                          **

@ 2,2 say 'placing data into transpose'
sele 1
 index on tube+str(section)+b->datecode to tube3
 go top

do while .not. eof()
  cur_tube=tube
  @ 3,2 say 'tube = '+cur_tube
 sele 5
  append blank
  replace tube with cur-tube
 sele 1
 do while cur_tube=tube
   @ 4,2 say 'section = '+str(section)+'  datecode = '+b->datecode
   m_name='dp'+alltrim(str(section))+'_dt'+alltrim(b->datecode)
   m_tube=tube
   m_RLD = str(turnover,7,3)
  sele 5
    replace &m_name with m_rld
  sele 1
   ship
 enddo
enddo
close all
```

```
** program transpose.prg - this program transposes minirhizotron data from a
* linear file to a file with each field representing a date depth combination.
* The first part of the program determines the scope and names
* of the fields (first part of the name is the section, the second part of
* the name is the date code). The second portion of the program places the
* data into the created database from the section database created by
* sectsum.prg.
* Written by Everett Weber on 9/14/94 last revision 9/14/94

close all
Clear

sele 2
use dates
index on ctod(month+'/'+day+'/'+year) to tube2

sele 3
 use structur
 zap
 append blank
  replace field_name with 'tube'
  replace field_type with 'C'
  replace field_len with 3
  replace field_dec with 0

* note fourth database will be opened called transpose
*    this database will contain the transposed data
sele 1
 use cluster
  set relation to date into b
  index on str(section)+b->datecode to tube unique
  go top
  clear
@ 2,2 say 'creating transpose'
do while .not. eof()
        m_name='dp'+alltrim(str(section))+'_dt'+alltrim(b->datecode)
        @ 3,2 say 'name = '+m_name
  sele 3
    append blank
    replace field_name with m_name
    replace field_type with 'N'
    replace field_len with 10
    replace field_dec with 3
```

```
  sele 1
        skip
 enddo

sele 5
create transp2 from structur
use transp2

*   end of file creation section                  **
** ---------------------------------------------------- **
** ---------------------------------------------------- **
*   section which transposes data                    **

@ 2,2 say 'placing data into transpose'
sele 1
 index on tube+str(section)+b->datecode to tube3
 go top

do while .not. eof()
  cur_tube=tube
 @ 3,2 say 'tube = '+cur_tube
  sele 5
  append blank
  replace tube with cur_tube
  sele 1
 do while cur_tube=tube
   @ 4,2 say 'section = '+str(section)+'  datecode = '+b->datecode
   m_name='dp'+alltrim(str(section))+'_dt'+alltrim(b->datecode)
   m_tube=tube
   m_RLD = rld
   sele 5
    replace &m_name with m_rld
   sele 1
   skip
 enddo
enddo
close all
```

```
** program trancoh.prg - this program transposes minirhizotron data from a
* linear file to a file with each field representing a date depth combination.
* The first part of the program determines the scope and names
* of the fields (first part of the name is the section, the second part of
* the name is the date code). The second portion of the program places the
* data into the created database from the section database created by
* sectsum.prg.
* Written by Everett Weber on 9/14/94 | last revision 9/14/94

close all
clear

sele 2
use dates
index on ctod(month+'/'+day+'/'+year) to tube2

sele 3
  use structur
  zap
  append blank
   replace field_name with 'tube'
   replace field_type with 'C'
   replace field_len with 3
   replace field_dec with 0
append blank
   replace field_name with 'init'
   replace field_type with 'n'
   replace field_len with 3
   replace field_dec with 0

* note fourth database will be opened called transpose
*    this database will contain the transposed data
sele 1
 use cohorfl
  set relation to date into b
  index on date to tube unique
  go top
  clear
@ 2,2 say 'creating transpose'
 do while not. eof()
         m_name='dt_'+alltrim(str(date,3,0))
         @ 3,2 say 'name = '+m_name
   sele 3
```

```
    append blank
    replace field_name with m_name
    replace field-type with 'c'
    replace field_len with 10
 * replace field_dec with 3
   sele 1
        skip
 enddo

sele 5
create trancoh from structur
use trancoh
clear
*   end of file creation section                    **
** ---------------------------------------------------- **
** ---------------------------------------------------- **
*  section which transposes data                      **

@ 2,2 say 'placing data into transpose'
sele 1
 index on tube+str(init,3,0)+str(date,3,0) to tube3
 go top

do while .not. eof()
  cur_tube=tube
  cur_init=init
  @ 3,2 say 'tube = '+cur_tube
  sele 5
  append blank
  replace tube with cur_tube
  replace init with cur_init
  sele 1
 do while cur_tube=tube and cur_init=init
   @ 4,2 say ' datecode = '+str(date,3,0)
   m_name='dt_'+alltrim(str(date,3,0))
   m_RLD = str(change, 10,5)
  sele 5
   replace &m_name with m_rld
  sele 1
   skip
 enddo
enddo
close all
```

```
* program tranbact.prg - program to retranspbse the file back to a linear format.
sele 2
use tranbac2
zap

sele 1
Use tranturn
go top

do while .not. eof()
        m_tube=tube
*       m_init=init
        m_p1t1=dp1_dt1
        m_p1t2=dp1_dt2
        m_p1t3=dp1_dt3
        m_p1t4=dp1_dt4
        m_p1t5=dp1_dt5
        m_p1t6=dp1_dt6
        m_p1t7=dp1_dt7
        m_p2t1=dp2_dt1
        m_p2t2=dp2_dt2
        m_p2t3=dp2_dt3
        m_p2t4=dp2_dt4
        m_p2t5=dp2_dt5
        m_p2t6=dp2_dt6
        m_p2t7=dp2_dt7
        m_p3t1=dp3_dt1
        m_p3t2=dp3_dt2
        m_p3t3=dp3_dt3
        m_p3t4=dp3_dt4
        m_p3t5=dp3_dt5
        m_p3t6=dp3_dt6
        m_p3t7=dp3_dt7

        sele 2
        append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 1
        replace depth with 1
        replace change with m_p1t1
        append blank
        replace tube with m_tube
```

```
*         replace init with m_init
          replace date with 2
          replace depth with 1
          replace change with m_p1 t2
     append blank
          replace tube with m_tube
*         replace init with m_init
          replace date with 3
          replace depth with I
          replace change with m_p1t3
        append blank
          replace tube with m_tube
*         replace init with m_init
          replace date with 4
          replace depth with 1
          replace change with m_p1t4
        append blank
          replace tube with m_tube
*         replace init with m_init
          replace date with 5
          replace depth with 1
          replace change with m_p1t5
     append blank
          replace tube with m_tube
*         replace init with m_init
          replace date with 6
          replace depth with 1
          replace changewith m_p1t6
        append blank
          replace tube with m_tube
*         replace init with m_init
          replace date with 7
          replace depth with 1
          replace change with m_p1 t7
        append blank
          replace tube with m_tube
*         replace init with m_init
          replace date with 1
          replace depth with 2
          replace change with m_p2t1
        append blank
          replace tube with m_tube
*         replace init with m_init
```

```
        replace date with 2
        replace depth with 2
        replace change with m_p2t2
    append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 3
        replace depth with 2
        replace change with m_p2t3
       append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 4
        replace depth with 2
        replace change with m_p2t4
       append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 5
        replace depth with 2
        replace change with m_p2t5
    append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 6
        replace depth with 2
        replace change with m_p2t6
       append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 7
        replace depth with 2
        replace change with m_p2t7
       append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 1
        replace depth with 3
        replace change with m_p3t 1
       append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 2
```

```
        replace depth with 3
        replace change with m_p3t2
    append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 3
        replace depth with 3
        replace change with m_p3t3
      append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 4
        replace depth with 3
        replace change with m_p3t4
      append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 5
        replace depth with 3
        replace change with m_p3t5
    append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 6
        replace depth with 3
        replace change with m_p3t6
      append blank
        replace tube with m_tube
*       replace init with m_init
        replace date with 7
        replace depth with 3
        replace change with m_p3t7
      sele 1
      skip
enddo
sele 2
replace change with str(val(change)/42,10,5) for date=1 and alltrim(change)<>'.'
replace change with str(val(change)/33,10,5) for date=2 and alltrim(change)<>'.'
replace change with str(val(change)/27,10,5) for date=3 and alltrim(change)<>'.'
replace change with str(val(change)/35,10,5) for date=4 and alltrim(change)<>'.'
replace change with str(val(change)/27,10,5) for date=5 and alltrim(change)<>'.'
replace change with str(val(change)/39,10,5) for date=6 and alltrim(change)<>'/'
replace change with str(val(change)/21,10,5) for date=7    and alltrim(change)<>'.'
```

```
* File: updat.prg -- program to update files; from roots
* first work area is file which contains the blank record
* hog contains the digitized data
* Note: don't forget to index data Wore browsing for errors
*      otherwise it appears that some records are missing
* written: 6/29/93 | updated: 7/25/94
sele 1
use output
        * data file with zero lengths
index on month++day+year+tube+frame to tube1
sele 2
use hog
        * data file which contains digitized info
index on month++day+year+tube+frame to tube2
sele 1
set relation to month++day+year+tube+frame into b
go top
do while .not. eof()
  ifb->length<>0 then
      replace length with b->length
      replace width with b->width
      replace root_id          with b->root_id
      replace seg_id with b->seg_id
      replace init with b->init
      section=arc_id
     search=month+day+year+tube+frame
      sele 2
      go top
      find &search
      m_month=month
      m-day-day
      m_year=year
      m_tube=tube
      m_frame=frame
      search2=month+m_day+m_year+tube+frame
      x=1
    do while search2=search
      sele 2
        go top
        find &search
        skip x
        m_length=length
        m_width=width
```

```
        m_init=init
        m_root_i=root_id
        m_seg_id=seg_id
        skip
        x=x+1
        search2=month++day+year+tube+frame
*suspend
       sele 1
        append blank
*suspend
        replace month with m_month
        replace day with m_day
        replace year with m_year
        replace tube with m_tube
        replace frame with m_frame
        replace length with m_length
        replace width with m_width
        replace init with m_init
        replace root_id with m_root_i
        replace seg_id with m_seg_id
        replace arc_id with section
*suspend
     enddo
     sele 1
  endif
  skip
enddo
close all
sele 2
 use frame
 index on tube+frame to frame
sele 1
use output
```

```
** program clustum.prg - this program sums all root within a specified
* section of the tube. Output from this file can then be used in other
* programs to find the RLD peak or other analyses. The program uses the
* file output.dbf as input and creates the file section.dbf.
* created 9/20/94 by Everett Weber; revised 10/16/94

close all

d e 2
use clusturn
zap


sele 3
use frame
index on tube+frame to tube2


sele 1
use turnover
set relation to tube+frame into c

dele for c->section>12 && eliminates sections greater than 12
dele for c->section<1
pack

index on dtoc(date)+tube+str(c->section) to tube
go top


do while .not. eof()
   sele 1
         m_tube=tube
         m_date=date
         m_sec=ceiling(c->section/4)
         M_RLD=0
         rootnumb=0
        do while tube=m_tube and m_date=date and m_sec=ceiling(c->section/4) and .not. eof()
         M_RLD=M_RLD+turnover
         rootnumb=rootnumb+1
         skip
        enddo
         if m_rld>0
```

```
        M_RLD=M_RLD/rootnumb
        endif
  sele 2
        append blank
        replace tube with m_tube
        replace date with m_date
        replace section with m_sec
        replace turnover with M_RLD
enddo
close all
```

```
** HOGI5.SAS Root length density sas program;

FILENAME REPEAT'TRANSP2 ASC Al';

OPTIONS LINESIZE = 75;
DATA NASAROOT;
   INFILE REPEAT;
   INPUT OBS TUBE
       DP1_DT1 DP1_DT2 DP1_DT3 DP1_DT4
       DP1_DT5 DP1_DT6 DP1_DT7 DP1_DT8
       DP2_DT1 DP2_DT2 DP2_DT3 DP2_DT4
       DP2_DT5 DP2_DT6 DP2_DT7 DP2_DT8
       DP3_DT1 DP3_DT2 DP3_DT3 DP3_DT4
       DP3_DT5 DP3_DT6 DP3_DT7 DP3_DT8;

       DP1_DT1 = LOG(DP1_DT1+1);
       DP1_DT2 = LOG(DP1_DT2+1);
       DP1_DT3 = LOG(DP1_DT3+1);
       DP1_DT4 = LOG(DP1_DT4+1);
       DP1_DT5 = LOG(DP1_DT5+1);
       DP1_DT6 = LOG(DP1_DT6+1);
       DP1_DT7 = LOG(DP1_DT7+1);
       DP1_DT8 = LOG(DP1_DT8+1);

       DP2_DT1 = LOG(DP2_DT1+1);
       DP2_DT2 = LOG(DP2_DT2+1);
       DP2_DT3 = LOG(DP2_DT3+1);
       DP2_DT4 = LOG(DP2_DT4+1);
       DP2_DT5 = LOG(DP2_DT5+1);
       DP2_DT6 = LOG(DP2_DT6+1);
       DP2_DT7 = LOG(DP2_DT7+1);
       DP2_DT8 = LOG(DP2_DT8+1);

       DP3_DT1 = LOG(DP3_DT1+1);
       DP3_DT2 = LOG(DP3_DT2+1);
       DP3_DT3 = LOG(DP3_DT3+1);
       DP3_DT4 = LOG(DP3_DT4+1);
       DP3_DT5 = LOG(DP3_DT5+1);
       DP3_DT6 = LOG(DP3_DT6+1);
       DP3_DT7 = LOG(DP3_DT7+1);
       DP3_DT8 = LOG(DP3_DT8+1);

* SETTING UP PLOTS;
```

```
IF T JBE=10 OR TUBE=14 OR TUBE=12 OR TUBE=1 1
  TI IEN PLOT=1;
IFT JBE=18 OR TUBE=25 OR CUBE-23 OR TUBE=2 1
  TI IEN PLOT=2;
IF T JBE=7 OR TUBE=13 OR TUBE=4 OR TUBE=1
  TI IEN PLOT=3;
IF T JBE=30 OR TUBE=36 OR TUBE=16 OR TUBE=31
  TI IEN PLOT=4;
IF T JBE=32 OR TUBE=35 OR TUBE=34 OR TUBE=33
  TI IEN PLOT=5;
IFT JBE=3 OR TUBE=5 OR TUBE=6 OR TUBE=8
  TI IEN PLOT=6;
IFT JBE=15 OR TUBE=9 OR TUBE=20 OR TUBE=2
  TI EN PLOT=7;
IFT JBE=19 OR TUBE=22 Ok TUBE=24 OR TUBE-17
  TI EN PLOT=8;

*ASSIGN VALUES OF 1 TO 4 TO TUBE2 FOR GLM PROCEDURE;
  IF TUBE = 10 OR TUBE = 7 OR TUBE = 30 OR TUBE = 32
    OR TUBE = 3 OR TUBE = 15 OR TUBE = 19 OR TUBE = 18
    THEN TUBE2 = 1;
  IF TUBE=14 OR TUBE=25 OR TUBE=13 OR TUBE=36 OR TUBE=35
    OR TUBE=5 OR TUBE=9 OR TUBE=22 THEN TUBE242;
  IF TUBE=12 OR TUBE=23 OR TUBE=4 OR TUBE=16 OR TUBE=34 OR
TUBE=6
    OR TUBE=20 OR TUBE=24 THEN TUBE2=3;
  IF TUBE=11 OR TUBE=21 OR TUBE=1 OR TUBE=3 1 OR TUBE=33 OR
TUBE=8
    OR TUBE=2 OR TUBE=17 THEN TUBE2=4;

* FERTILIZATION;
  IF PLOT=1 OR PLOT=3 OR PLOT=4 OR PLOT=5
    THEN TREAT='FERTILIZED';
  IF PLOT=2 OR PLOT=6 OR PLOT=7 OR PLOT=8
    THEN TREAT='UNFERTILIZED';

* SPECIAL ADDITIONS FOR THIS RUN;
  * PLOT 2 WAS A CLEAR OUTLYER;
IF PLOT=2 THEN DELETE;

*  REMOVING UNWANTED TREATMENTS;

*                              ,
```

```
*        END OF DATA STEP
*-----------------------------------------------------;
PROC SORT;
  BY TREAT PLOT TUBE2;

PROC GLM ;
  CLASS TREAT PLOT TUBE2;
  MODEL
        DP1_DT1 DP1_DT2 DP1_DT3 DP1_DT4
        DP1_DT5 DP1_DT6 DP1_DT7 DP1_DT8
        DP2_DT1 DP2_DT2 DP2_DT3 DP2_DT4
        DP2_DT5 DP2_DT6 DP2_DT7 DP2_DT8
        DP3_DT1 DP3_DT2 DP3_DT3 DP3_DT4
        DP3_DT5 DP3_DT6 DP3_DT7 DP3_DT8
        = TREAT PLOT(TREAT)
        / SS4 NOUNI;
  REPEATED DEPTH 3, DATE 8 / NOM PRINTE;
RUN;
ENDSAS;
```

```
* * HOGITUR3. SAS  Turnover sas program;

FILENAME REPEAT 'TURNMISS ASC Al';

OPTIONS LINESIZE = 75;
DATA NASAROOT;
   INFILE REPEAT;
   INPUT OBS TUBE
       DP1_DT1 DP1_DT2 DP1_DT3 DP1_DT4
       DP1_DT5 DP1_DT6 DP1_DT7
       DP2_DT1 DP2_DT2 DP2_DT3 DP2_DT4
       DP2_DT5 DP2_DT6 DP2_DT7
       DP3_DT1 DP3_DT2 DP3_DT3 DP3_DT4
       DP3_DT5 DP3_DT6 DP3_DT7       ;

*        IF DP1_DT1 = . THEN DP1_DT1 = .6157;
*        IF DP1_DT2 = . THEN DP1_DT2 = .6157;
*        IF DP1_DT3 = . THEN DP1_DT3 = .6157;
*        IF DP1_DT4 = . THEN DP1_DT4 = .6157;
*        IF DP1_DT5 = . THEN DP1_DT5 = .6157;
*        IF DP1_DT6 = . THEN DP1_DT6 = .6157;
*        IF DP1_DT7 = . THEN DP1_DT7 = .6157;

*        IF DP2_DT1 = . THEN DP2_DT1 = .6157;
*        IF DP2_DT2 = . THEN DP2_DT2 = .6157;
*        IF DP2_DT3 = . THEN DP2_DT3 = .6157;
*        IF DP2_DT4 = . THEN DP2_DT4 = .6157;
*        IF DP2_DT5 = . THEN DP2_DT5 = .6157;
*        IF DP2_DT6 = . THEN DP2_DT6 = .6157;
*        IF DP2_DT7 = . THEN DP2_DT7 = .6157;

*        IF DP3_DT1 = . THEN DP3_DT1 = .6157;
*        IF DP3_DT2 = . THEN DP3_DT2 = .6157;
*        IF DP3_DT3 = . THEN DP3_DT3 = .6157;
*        IF DP3_DT4 = . THEN DP3_DT4 = .6157;
*        IF DP3_DT5 = . THEN DP3_DT5 = .6157;
*        IF DP3_DT6 = . THEN DP3_DT6 = .6157;
*        IF DP3_DT7 = , THEN DP3_DT7 = .6157;
* CREATING THE CORRECT TURNOVER DATA BY PROVIDING A RATE
(TURNOVER/DAY);
 DP1_DT1 = DP1_DT1/42;
 DP1_DT2 = DP1_DT2/33;
```

```
DP1_DT3 = DP1_DT3/27;
DP1_DT4 = DP1_DT4/35;
DP1_DT5 = DP1_DT5/27;
DP1_DT6 = DP1_DT6/39;
DP1_DT7 = DP1_DT7/21;

DP2_DT1 = DP2_DT1/42;
DP2_DT2 = DP2_DT2/33;
DP2_DT3 = DP2_DT3/27;
DP2_DT4 = DP2_DT4/35;
DP2_DT5 = DP2_DT5/27;
DP2_DT6 = DP2_DT6/39;
DP2_DT7 = DP2_DT7/21;

DP3_DT1 = DP3_DT1/42;
DP3_DT2 = DP3_DT2/33;
DP3_DT3 = DP3_DT3/27;
DP3_DT4 = DP3_DT4/35;
DP3_DT5 = DP3_DT5/27;
DP3_DT6 = DP3_DT6/39;
DP3_DT7 = DP3_DT7/21;

    DP1_DT1 = ARSIN(DP1_DT1);
    DP1_DT2 = ARSIN(DP1_DT2);
    DP1_DT3 = ARSIN(DP1_DT3);
    DP1_DT4 = ARSIN(DP1_DT4);
    DP1_DT5 = ARSIN(DP1_DT5);
    DP1_DT6 = ARSIN(DP1_DT6);
    DP1_DT7 = ARSIN(DP1_DT7);

    DP2_DT1 = ARSIN(DP2_DT1);
    DP2_DT2 = ARSIN(DP2_DT2);
    DP2_DT3 = ARSIN(DP2_DT3);
    DP2_DT4 = ARSIN(DP2_DT4);
    DP2_DT5 = ARSIN(DP2_DT5);
    DP2_DT6 = ARSIN(DP2_DT6);
    DP2_DT7 = ARSIN(DP2_DT7);

    DP3_DT1 = ARSIN(DP3_DT1);
    DP3_DT2 = ARSIN(DP3_DT2);
    DP3_DT3 = ARSIN(DP3_DT3);
    DP3_DT4 = ARSIN(DP3_DT4);
    DP3_DT5 = ARSIN(DP3_DT5);
```

```
        DP3_DT6 = ARSIN(DP3_DT6);
        DP3_DT7 = ARSIN(DP3_DT7);

* SETTING UP PLOTS;
   IF TUBE=10 OR TUBE=14 OR TUBE=12 OR TUBE=11
     THEN PLOT=1;
   IF TUBE=18 OR TUBE=25 OR TUBE=23 OR TUBE-21
     THEN PLOT-IL;
   IF TUBE=7 OR TUBE=13 OR TUBE=4 OR TUBE-1
     THEN PLOT-3;
   IF TUBE=30 OR TUBE==36 OR TUBE=16 OR TUBE==3 1
     THEN PLOT=4;
   IF TUBE=32 OR TUBE=35 OR TUBE=34 OR TUBE=33
     THEN PLOT=5;
   IF TUBE=3 OR TUBE=5 OR TUBE=6 OR TUBE-8
     THEN PLOT=6;
   IF TUBE=15 OR TUBE=9 OR TUBE=20 OR TUBE=2
     THEN PLOT=7;
   IF TUBE=19 OR TUBE=22 OR TUBE=24 OR TUBE=17
     THEN PLOT=8;

*ASSIGN VALUES OF 1 TO 4 TO TUBE2 FOR GLM PROCEDURE;
   IF TUBE = 10 OR TUBE = 7 OR TUBE = 30 OR TUBE = 32
     OR TUBE = 3 OR TUBE = 15 OR TUBE = 19 OR TUBE = 18
     THEN TUBE2 = 1;
   IF TUBE=14 OR TUBE=25 OR TUBE=13 OR TUBE=36 OR TUBE=35
     OR TUBE=5 OR TUBE=9 OR TUBE=22 THEN TUBE2=2;
   IF TUBE=12 OR TUBE=23 OR TUBE=4 OR TUBE=16 OR TUBE==34 OR
TUBE=6
     OR TUBE=20 OR TUBE=24 THEN TUBE2=3;
   IF TUBE=11 OR TUBE=21 OR TUBE=1 OR TUBE=3 1 OR TUBE-33 OR
TUBE=8
     OR TUBE=2 OR TUBE=17 THEN TUBE2=4;

* FERTILIZATION;
   IF PLOT=1 OR PLOT=3 OR PLOT=4 OR PLOT=5
     THEN TREAT='FERTILIZED';
   IF PLOT=2 OR PLOT=6 OR PLOT-7 OR PLOT=%
     THEN TREAT='UNFERTILIZED';

* SPECIAL ADDITIONS FOR THIS RUN;
  * PLOT 2 WAS A CLEAR OUTLYER;
IF PLOT=2 THEN DELETE;
```

```
* REMOVINGUNWANTED TREATMENTS;

*                              .      ,
*      E N D O F D A T A S T E P       ;
*-------------------------------------------;
PROC SORT;
 BY TREAT PLOT TUBE2;
PROC PRINT;
PROC GLM ;
 CLASS TREAT PLOT TUBE2;
 MODEL

      DP1_DT5 DP1_DT6 DP1_DT7

      DP2_DT5 DP2_DT6 DP2_DT7

      DP3_DT5 DP3_DT6 DP3_DT7
      = TREAT PLOT(TREAT)
      / ss4 NOUNI;
 REPEATED DEPTH 3, DATE 3 / NOM PRINTE;
 MEAN DATE/TUKEY REGWF;
 MEAN DEPTH*TREAT/TUKEY REGWF;
RUN;
ENDSAS;
```

```
** COHORT.SAS Cohort analysis sas program;

FILENAME COHORT 'TRANCOH TXT A1';
FILENAME TURNDATE 'TURNDATE ASC Al';
OPTIONS LINESIZE=75;
DATA TURN;
  INFILE COHORT;
  INPUT OBS TUBE INIT
       DT_1 DT_2 DT_3 DT_4;

* SETTING UP PLOTS;
   IF TUBE-10 OR TUBE-14 OR TUBE-12 OR TUBE=1 1
     THEN PLOT=1;
   IF TUBE-18 OR TUBE=25 OR TUBE=23 OR TUBE-21
     THEN PLOT-2;
   IF TUBE-7 OR TUBE-13 OR TUBE=4 OR TUBE=1
     THEN PLOT=3;
   IF TUBE-30 OR TUBE=36 OR TUBE-16 OR TUBE-31
     THEN PLOT4;
   IF TUBE=32 OR TUBE-35 OR TUBE=34 OR TUBE=33
     THEN PLOT=5;
   IF TUBE-3 OR TUBE=5 OR TUBE=6 OR TUBE=8
     THEN PLOT=6;
   IF TUBE=15 OR TUBE=9 0R TUBE=20 OR TUBE=2
     THEN PLOT=7;
   IF TUBE=19 OR TUBE==22 OR TUBE-24 OR TUBE=17
     THEN PLOT=8;

* FERTILIZATION;
   IF PLOT=1 OR PLOT=3 OR PLOT=4 OR PLOT=5
     THEN TREAT='F';
   IF PLOT=2 OR PLOT=6 OR PLOT=7 OR PLOT==8
     THEN TREAT='U';
* REMOVAL OF UNWANTED DATA;
  IF PLOT=2 THEN DELETE;
* ADJUSTING DATA TO DATE

* IF INIT=3 THEN DT_1=DT_1/27;
* IF INIT=3 THEN DT_2=DT_2/35;
* IF INIT=3 THEN DT_3=DT_3/27;
* IF INIT=4 THEN DT_1=DT_1/35 ;
* IF INIT=4 THEN DT_2=DT_2/27;
```

```
* IF INIT=4 THEN DT_3=DT_3/39;
* IF INIT=5 THEN DT_1=DT_1/27;
* IF INIT=5 THEN DT_2=DT_2/39;
* IF INIT=5 THEN DT_3=DT_3/21;

PROC GLM;
    CLASS TREAT PLOT INIT;
    MODEL DT_1 DT_2 DT_3 = TREAT PLOT(TREAT) INIT TREAT*INIT /SS4;
    REPEATED DATE 3 POLYNOMIAL/SUMMARY,

RUN;
ENDSAS;
```