EFFECTS OF SOIL COMPACTION AND ORGANIC MATTER REMOVAL ON MORPHOLOGY OF SECONDARY ROOTS OF LOBLOLLY PINE

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Abstract—Root studies are being used to monitor possible changes in growth of loblolly pines on a long-term soil productivity study site. Here, we report the results of a preliminary look at roots in the sixth growing season. Roots were collected from loblolly pines grown in soil that was first subjected to three levels of compaction (none, moderate, severe) and three levels of organic matter removal (stem only, total tree, and total aboveground biomass). Roots were fixed, sectioned, and stained for examination by light microscope. The proportion of roots with bark formation decreased from 70 percent in uncompacted soil to 43 percent in severely compacted soil. Depletion of starch grains was significantly less in samples from uncompacted soil than in compacted soil.

INTRODUCTION

Management practices that change soil properties may affect growth and health of secondary roots and eventually the long-term productivity of the site. The USDA Forest Service's long-term soil productivity (LTSP) study, which has recently been installed at several sites in the United States and Canada (Tiarks and others 1993) is designed to measure such changes over a rotation. Its primary purpose is to monitor changes in productivity and soil processes. Three levels of soil compaction, three levels of organic matter removal, and two levels of vegetation control were applied (Powers and others 1990). Ultimately, interpretation of the relationship between these treatments and productivity will require the linking of changes in soil properties to soil processes, including root growth and health.

Anatomical study of roots is particularly useful for assessing root health (Walkinshaw 1995). Soil compaction affects root health and subsequent crop production in annual crops (Allmaras and others 1988, Feldman 1984). Compacted soils have fewer and smaller pores, and such conditions damage roots and alter their morphology. Loblolly pine roots less than about 5 millimeter (mm) in diameter appear to be particularly vulnerable to soil compaction (Copeland 1952). Shedding of dead cortex reduces root diameter and eliminates large numbers of root hairs in loblolly and other conifers (Kozlowski and Scholtes 1948, Leshem 1974). Many injuries develop after the period of root extension during secondary root development (Coutts 1987).

The objectives of the present study were to evaluate the usefulness of root morphology variables as indicators of root health, and to measure the effects of soil compaction and organic residue removal on anatomy of secondary roots.

METHODS

The root samples used were collected at the LTSP site located in Rapides Parish, LA, (Tiarks and others 1991) at the beginning and end of the sixth growing season. A 40-year-old loblolly pine stand on the site was harvested in 1989. The compaction treatments (none, moderate, and severe) and organic matter treatments (stem only, total tree, and total aboveground biomass) were applied soon after harvest. The plots were planted with containerized seedlings from 10 open-pollinated loblolly pine families in February 1990. Soil bulk densities were measured with a core sampler at planting and at stand age 5. Tree volumes were calculated on the basis of pine heights and diameter at breast height (d.b.h.) measured at age 6 (Schmitt and Bower 1970).

Roots were sampled twice in the sixth growing season. In the first sampling (March 24, 1995) roots from 10 trees were collected in the plots representing the extremes of the treatments (OM_0C_0 and OM_2C_2). In the second sampling (October 10, 1995) we sampled roots of five pines per treatment. There were 9 different treatments, and an average of 14 roots per tree was collected at each sampling. The March sampling yielded 264 root specimens and the October sampling 753. Root samples were from pines randomly selected from among the families. To ensure that nonpine roots were not included in the samples, we collected only in subplots that had received herbicide treatment.

Roots were collected in a 25-centimeter (cm) by 25-cm area located about 1 meter (m) from the stem of a pine. All soil and roots between a depth of 2 cm and a depth of 20 cm were collected and the roots gently shaken to remove excess soil. The center 2 to 4 cm of each root was excised and placed into formalin-acetic acid-alcohol (FAA) fixative for 14 days (Sass 1951). Fixed root specimens were re-cut to 1 to 3 mm, dehydrated in alcohol series, embedded in paraffin, and cut into 7 micrometers (m) to 10 m transverse sections. Sections were stained by hematoxylin-eosin, Papanicolaou's schedule, or the acid-schiff procedure (Haas 1980). Three to nine stained sections were prepared from each root, but only the first usable section from each root was read. Observations on root sections were at 100 to 500 diameters with a photomicroscope. Halogen and polarized light sources and neutral density filters were used

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to differentiate tannin and cellulose:lignin complexes. The percentage of roots meeting the criteria for a variable was calculated by tree. If the cambium appeared dead and tannin accumulation was excessive, the root was considered to be dead and not used in further analysis. No dead roots were found in the March sampling and only four roots were dead in the October sampling.

Variables for Histological Observations

Based on past observations of root samples from Louisiana (2,619 roots), Mississippi (623 roots), and North Carolina (1,221 roots), 10 variables were selected for further testing. They were:

Variable	Description
Cortex shedding	Cortical cells are dead and remain attached or are released into the soil. The shed is invaded by many soil microbes.
Periderm formation	The first stage in replacement of shed cortex. Cortical cells are coated with a protective layer of tannin and cellulose:lignin complex.
Bark formation	Intact layer of bark cells identical to those found in the stem encompasses the root. Protects the root from injuries and microbial invasion.
Starch	Degradation of starch grains in the cells is degradation 50 percent in the cells. Starch test in cortical and ray cells is negative.
Tannin	Accumulation of tannin-containing cells in accumulation the cortex, rays, and inner xylem. Number of cells with accumulations range from less than 10 to over 100.
Mycorrhizal	Status of short roots that are shed during status the early stage of cortical-cell death. New lateral roots often develop at the sites where mycorrhizal roots have shed.
Lateral root	Formation of roots that arise from the xylem formation and phloem and remain permanent rather than being shed like root hairs and mycorrhizae.
Pathogenic	Infection of living cambial or fiber cells, infection in contrast to invasion of the dead cortex in the shed. Root appears to die from this invasion.
Section	Tissue tearing that occurs during sectioning tearing and makes it difficult to examine specimens for periderm formation, starch degradation, and formation of lateral roots.
Number of	Number of starch-containing plastids per cell starch grains when the cell is viewed at a single focal per cell length at 100 to 500 diameters.

Data Analysis

Treatments were not replicated, but the plots were sufficiently large and uniform so that within-plot sampling could be considered replication in this preliminary test of root variables. Thus, trees within plots were used as replications. Analysis of variance was used to separate the means of the two treatments sampled in March. For the October sampling, analysis of variance for a factorial design was used to test the significance of the main effects (compaction and organic matter level) and of their interaction.

Results and Discussion

March sampling—Four of the 10 variables measured on samples collected March 24, 1995, were significantly affected by the two soil treatments (table 1). The combination of soil compaction and removal of aboveground organic matter delayed cortex shedding, periderm formation, and starch degradation. The proportion of roots with complete bark was 70 percent in the severely treated plot and only 43 percent in the low-impact

Table 1—Measurements of root samples collected March 24, 1995 by soil treatments

	Compaction-OM treatment		
Root variable	None-stem removal only	Severe-total aboveground	Probability of> F value
	Percentage of roots		
Cortex shedding	52	31	0.012
Periderm formation	53	28	0.002
Bark formation	43	70	0.001
Starch degradation	29	6	0.001
Tannin accumulation	12	12	0.830
Mycorrhizal status	9	8	0.851
Lateral root formation	6	5	0.809
Section tearing	28	18	0.166
Pathogenic infection	0	0	1.000
	Grains per cell		
Starch formation	10.9	10.7	0.848

treatment. The other variables were not significantly affected by the soil treatments.

October sampling—Of the 10 variables measured in the fall sampling, only 4 were significantly affected by the treatments (table 2). Cortex shedding, periderm formation, section tearing, and number of starch grains per cell were affected by soil compaction, while only periderm formation and number of starch grains per cell were significantly affected by removal of organic matter. None of the interaction terms was significant.

Removal of limbs and foliage during the harvest decreased the proportion of roots with periderm formation from 35 to 24 percent (table 3). Removal of the forest floor had no further effect on the percentage of roots with periderm formation. Removal of organic matter had a mixed effect on the number of starch grains per root cell, with the greatest number of starch grains per cell occurring in samples from the intermediate treatment. Although this effect was statistically significant, a biological explanation is not readily apparent.

As compaction increased, the percentage of roots with periderm formation decreased (table 3), with the greatest effect occurring as compaction was increased from 1.40 to 1.46 grams per cubic centimeter. The number of starch grains per root cell increased as the level of compaction increased. The change in number of starch grains per cell was proportional to the change in bulk density. The percentage of roots with cortex shedding decreased as

Table 2—Probability of differences occurring by chance for root variables in samples collected October 10, 1995

Root variable	Compaction	Organic matter treatment	Interaction
	Probability		
Cortex shedding	0.017	0.164	0.060
Periderm formation	0.016	0.045	0.171
Bark formation	0.084	0.126	0.299
Starch degradation	1.00[a]	1.00	1.00
Tannin accumulation	0.362	0.466	0.132
Mycorrhizal status	0.357	0.145	0.300
Lateral root formation	0.556	0.128	0.168
Section tearing	0.030	0.577	0.978
Pathogenic infection	0.289	0.537	0.586
Starch grains	0.001	0.025	0.089

[a] Only four roots showed signs of starch degradation.

Table 3—Means (percentage) of roots with periderm forming and number of starch grains per root cell, by soil treatment, for sample collected October 10, 1995

Compaction	Organic matter removed: Stem only	Total tree	Total above ground	Mean
	Pct. of roots with periderm forming			
None (1.40a)	45	27	33	65
Moderate (1.46)	28	27	31	27
Severe (1.49)	33	20	9	21
Mean	35	24	25	
	Number of starch grains per cell			
None (1.40)	5.0	6.7	6.1	5.9
Moderate (1.46)	7.0	6.9	6.4	6.7
Severe (1.49)	6.8	7.5	7.1	7.1
Mean	6.3	7.0	6.5	

a = bulk density (grams per cubic centimeter) at age 5.

bulk density increased from 1.40 to 1.46 grams per cubic centimeter but was not affected by a further increase in compaction (table 4). The percentage of sections that were torn decreased as the compaction level decreased. Percentage of torn sections could be an artifact of the sampling or related to the physical strength of the roots. Further evaluation of this variable will be necessary if the meaning of the difference is to be understood.

The compaction and organic-matter removal treatments affected pine height and volume growth by age 6 (table 5). In general, the plot means for volume declined with increased compaction. The same was true for percentage of roots with cortex shedding and periderm formation. When the height and volume of single trees were regressed against root variables for the same trees, no significant relationships were apparent. However, the sampling method does not guarantee that a root sample is from the correct tree.

Table 4—Effect of soil compaction on percentage of roots with cortex shedding and torn sections

Compaction	Bulk density	Cortex shedding	Torn section
	Grams/cm^{-3}	*Percentage*	
None	1.40	52	20
Moderate	1.46	38	18
Severe	1.49	38	10

Table 5—Effect of compaction and organic matter removal on growth of loblolly pine at age 6

Organic matter removal	Compaction treatment	Soil bulk density	Height	Volume
	G/cm^{3}	*m*	*D*	m^3/tree
Stem only	None	1.40	5.6	15.8
	Moderate	1.46	5.9	18.3
	Severe	1.49	4.8	9.5
Total tree	None	1.40	4.7	9.2
	Moderate	1.46	5.0	11.0
	Severe	1.49	4.9	11.0
Total aboveground	None	1.40	4.9	9.6
	Moderate	1.46	5.1	10.9
	Severe	1.49	4.4	7.8

Tannin accumulation, pathogenic infection, mycorrhizal status, and formation of lateral roots were unaffected by the soil treatments. These variables have shown promise as indicators of root health in older stands. In this 6-year-old stand, the roots were all generally healthy. Root morphological characteristics that were affected by the compaction, such as periderm formation and number of starch grains per cell, are probably more indicative of tree vigor than of the health of root systems.

CONCLUSIONS

A variety of histological measurements used in the two root samplings significantly separated effects of soil treatments. The variables we measured were easy to tabulate and had relatively low coefficients of variation. Overall, the root health of the trees growing in all soil treatments appeared the same. Cortex shedding and periderm formation, which may be predictors of pine growth, were the only two variables statistically affected by the soil treatments in both sampling periods. Degradation of starch and formation of bark were significantly different by treatment only in the March sampling, while number of starch grains per cell differed by treatment in the October sampling. Soil compaction had a more consistent effect on root morphology than did organic matter removal.

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