CHAPTER III

METHODS AND MATERIALS

Site and Soil Descriptions

The four soils chosen for this study, Dome, Cohasset, Clarksville and Argent series, represent contrasting forest soils from LTSP field sites across the United States. All four soils are of moderate to large extent in their region and are important for timber production. The Dome and Cohasset soils come from sites in the California Sierra Nevada Range, while the Clarksville soil is from a site in the Missouri Ozark Range, and Argent is from a South Carolina Lower Coastal Plain site. Typical profile horizonation for each soil is presented in Fig. III.1.

The Dome, Cohasset and Clarksville soils were taken from established LTSP research sites, while the Argent soil was taken from a Mead-Westvaco/Virginia Tech long-term productivity research site. The Dome site is located on the Sierra National Forest, Madera County, California at an elevation of 1576 m. The Dome series are Typic Dystroxerrepts developed from granodiorite of the mesozoic Sierra Nevada Batholith. In contrast, Cohasset soils are Ultic Haploxeralfs formed from cenozoic volcanic andesite mudflows. Cohasset soil was taken from the Blodgett Experimental Forest LTSP study site in El Dorado County, California, at an elevation of 1350 m. Clarksville soil was taken from Carr Creek State Forest, Shannon County, Missouri. This Typic Paleudult developed from Ordovician and Cambrian dolomites, cherty dolomites and some sandstones of the Gasconade formation (Gott, 1975). Argent soil was taken from a lower coastal plain site in Colleton County, South Carolina. The Argent series are Typic Endoaqualfs developed on relatively flat terrain from marine sediments in a subtropical climate.

Surface soil, taken from the 0 to 20 cm depth, was used in this experiment. This depth encompassed all or most of each soil's surface A horizon. Soil samples were air dried and sieved (2 mm) to obtain the fine-earth fraction. Particle size analysis was done for each soil by first oxidizing organic matter (Gee and Bauder, 1986), performing

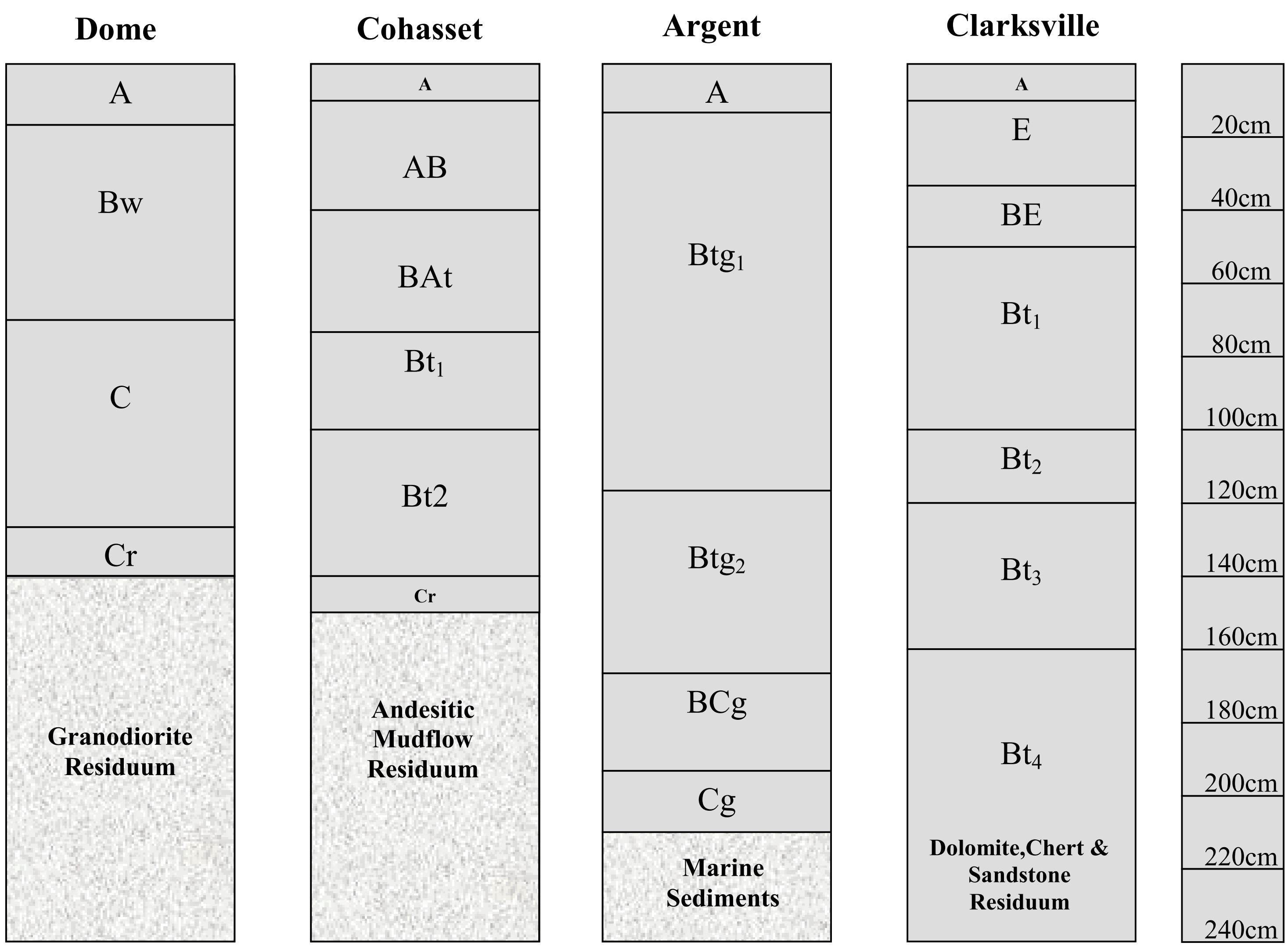

Figure III.1. Profile comparison of the four forest soils from various forest regions used in this study. The Dome and Cohasset soils are from California, the Argent from South Carolina, and the Clarksville from Missouri.

standard mechanical analysis (ASTM, 1972), and then wet-sieving and sand fractionation. Carbon and nitrogen were determined by a combustion method using a vario MAX CNS analyzer (Elementar, Hanau, Germany). Organic matter percent was determined as the organic carbon content multiplied by 1.72 (Nelson and Sommers, 1982).

Experimental Design and Layout Overview

A seven by seven (ρ_b by θ_v) factorial greenhouse experiment was performed to assess root growth as a function of soil bulk density and soil volumetric water content (Fig. III.2). A series of soil compaction tests determined the optimum technique for uniformly compacting soils in PVC cylinders that were used to assess compaction effects on various soil physical properties and to grow tree seedlings. PVC cylinders with dimensions of 8 cm x 15 cm were packed at seven bulk density levels with surface soil from each LTSP site. Seeds from tree species associated with each soil type were grown and planted in the soil pots. Pots with soil from each of the seven ρ_b levels were maintained at seven different θ_v throughout the growing period. Seedling root and shoot growth were measured at the end of the growth period. Multiple regression analyses were performed to develop **r**esponse surfaces describing seedling growth as a function of density and water. In addition, LLWR was determined for each soil and the effect of LLWR on growth analyzed.

Soil Compaction

Compaction Equipment

Compaction testing equipment was manufactured specifically for this experiment to meet ASTM standards and allow us to use standard 8 cm PVC pipe for compacting and testing soils and as pots for planted seedlings (ASTM, 1996). A slide hammer was manufactured to meet weight specifications (2.5 kg) and slide smoothly in the PVC cylinder. A metal base with the same diameter as the PVC cylinder attached to a metal rod was designed to sit on the soil surface. The compaction hammer slid down the rod

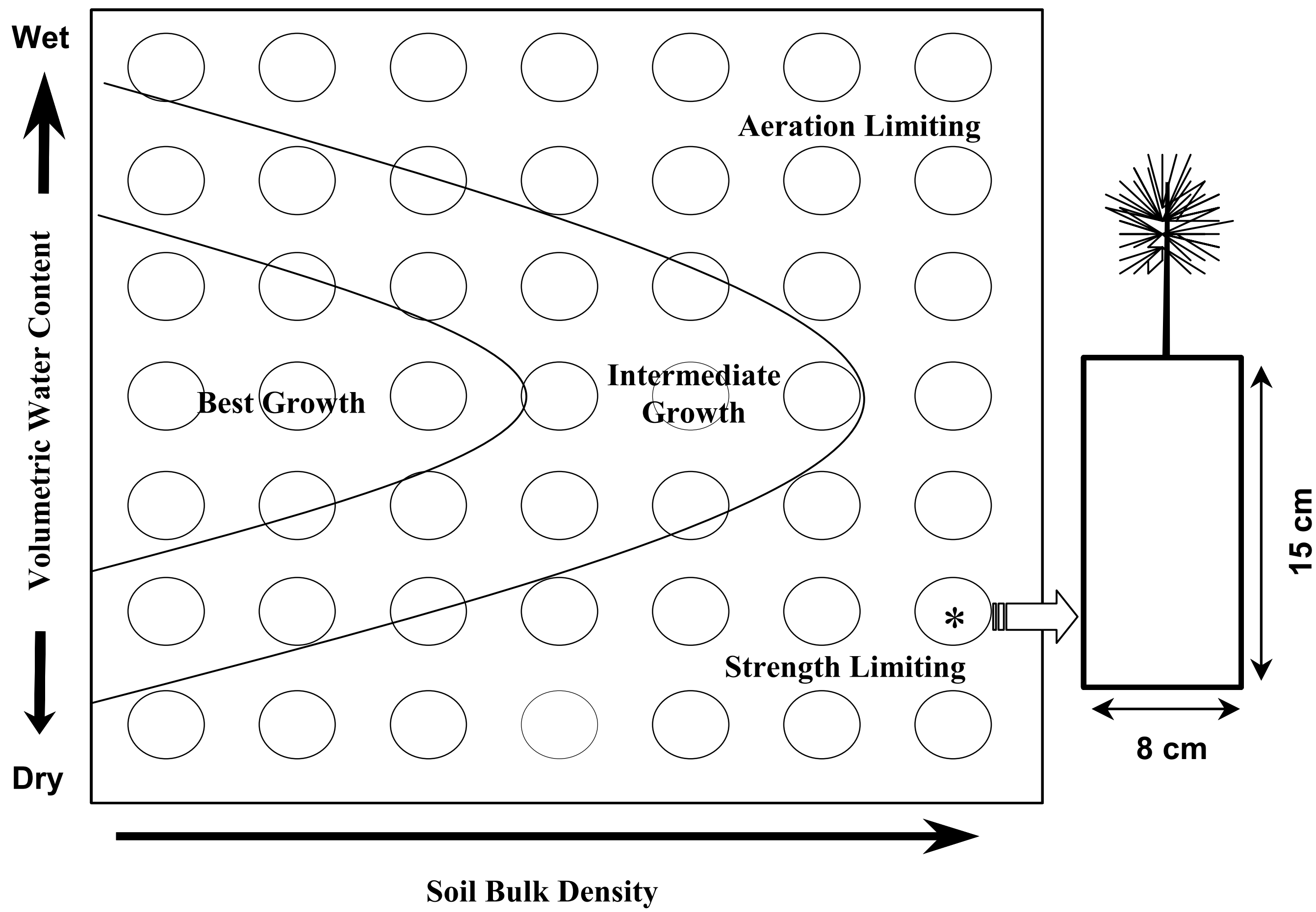

Figure III.2. Experimental layout of a greenhouse experiment to determine root growth opportunity as a function of soil volumetric water content and bulk density for four forest soils and tree species. Each circle represents a PVC pot with planted seedling.

allowing each hammer blow to evenly compact the soil layer. In addition, a brace was manufactured to secure the cylinder and hammer during soil compaction thus allowing the hammer to drop consistently from 30.5 cm and provide uniform compaction. For this study, we defined a "hammer blow" as the drop of the compaction hammer from 30.45 cm onto the metal plate covering the soil in the PVC cylinder, thus compacting the soil. Compactive effort (kN m^{-3}) is the force associated with each hammer blow.

We tested our compaction equipment to determine if it would uniformly compact the soil in the PVC cylinders. Argent soil at a uniform water content was compacted in 4 lifts in a 15 cm tall PVC cylinder. Soil was added loosely to a depth of 5 cm and five hammer blows were applied to each 5-cm lift. The soil column was carefully cut into four segments using a masonry saw. Soil dimensions and mass were measured, the cores oven dried, and ρ_b determined. The ρ_b of the four lifts, from top to bottom, was 1.46, 1.52, 1.52, and 1.53 Mg m^{-3} showing that we could achieve relatively uniform compaction within the experimental PVC cylinders.

Optimum Water Content

Soil compaction standard effort tests (also called the Proctor test) of each soil determined the OWC for compaction and maximum dry density (ASTM, 1996). Optimum water content is defined as the water content at which a soil reaches maximum density in laboratory tests (ASTM, 1996). Literature-based target values were used to estimate each soil's OWC. Air-dry soil was brought to five different water contents, bracketing the literature-based estimate. Water was slowly added as the soil turned in a mechanical mixer. Soil cohesiveness and aggregate size were used to estimate the spectrum of water content. The moistened soil was covered and allowed to equilibrate for 24 hours to ensure uniform soil wetness. This procedure was effective for bracketing the target moisture content and creating well-mixed, uniformly moist soil. Soil was sampled and oven-dried to determine gravimetric water content.

Replicate cores for each soil water content level were compacted in 8 by 10 cm PVC cylinders. Loose soil was added, the surface settled and smoothed, and then compacted in two 5-cm lifts with 25 hammer blows per lift. Soil volume, mass, and water content were measured and oven dry weight and ρ_b determined for each core.

Optimum water content was determined from graphs plotting water content as a function of ρ_b for each soil type. Subsequently, all soils were compacted at their OWC.

Compactive Effort

Compactive effort for each soil was determined as a variation of the ASTM compaction standard effort tests to assess differences in each soil's ρ_b range and compactibility. Also, this test determined the compactive effort (number of hammer blows) needed to achieve target ρ_b for compacted soil cores used in subsequent analyses. Soil was moistened at the OWC and then compacted in replicate PVC cylinders as described previously. Each core received a set number of blows (1, 2, 4, 8, 16, 32, and 64 blows) to correlate a range of compaction hammer blows and ρ_b, similar to the work done by Howard et al. (1981). The compactive effort associated with each level of hammer blows was determined. Minimum and maximum densities for each soil were determined from graphs depicting ρ_b as a function of compactive effort, with maximum density being defined as the asymptote of the curve.

Compression Index

The relationship of ρ_b increasing with increasing standard effort is similar to that found for compression curves developed for soils measured in one-dimensional consolidation tests, where ρ_b increases (or void ratio decreases) as a function of the log of applied stress (Larson et al., 1980; McNabb and Boersma, 1993). Although testing procedures and forces applied differed, we used these principals to test compactibility and determine the compression index (CI) of our soils. CI is defined as the slope of the linear portion of a compression curve (Larson et al., 1980; McNabb and Boersma, 1993). The CI is an indicator of a soil's compressibility. A lower CI indicates that the rate of ρ_b increase with applied stress is less than that of a soil with a higher CI. The regression equations defining each soil's compression index were determined and tested using dummy variables to detect slope differences, and thus compressibility, between the soils.

Soil Bulk Density

Regression analyses on transformed data were used to determine the relationship between compactive effort and ρ_b for each soil (Table III.1). Seven compaction levels for each soil were determined using the minimum and maximum densities and the regression equation describing ρ_b as a function of compaction hammer blows. Forty-nine soil columns for each soil type were constructed. Bulk quantities of each soil were brought to its OWC. Seven PVC cylinders for each density level were filled and compacted using procedures previously described. Gravimetric water content samples of each soil were periodically collected during the soil compaction procedure to ensure consistent soil water content. Bulk density of all soil columns was determined after compaction (Table III.2).

Table III.1. Regression equations and calculated values to determine the number of compaction hammer blows needed to achieve target bulk densities for four forest soils.

Dome		Cohasset		Clarksville		Argent	
$\log y^2 = 0.1154 * \log x^2 + .0276$, $r^2 = 0.99$		$\log y^2 = 0.1049 * \log x^2 - 0.037$, $r^2 = 0.95$		$\log y^2 = 0.0964 * \log x^2 + 0.141$, $r^2 = 0.95$		$\log y^2 = 0.0823 * \log x^2 + 0.197$, $r^2 = 0.95$	
Target Bulk Density (Mg m^{-3})	Calculated No. Blows	Target Bulk Density (Mg m^{-3})	Calculated No. Blows	Target Bulk Density (Mg m^{-3})	Calculated No. Blows	Target Bulk Density (Mg m^{-3})	Calculated No. Blows
1.04	1	0.94	1	1.16	1	1.24	1
1.12	2	1.01	2	1.23	2	1.31	2
1.20	4	1.07	3	1.30	3	1.39	3
1.28	6	1.14	5	1.37	5	1.46	6
1.36	11	1.20	9	1.44	8	1.53	9
1.44	18	1.27	14	1.51	13	1.61	21
1.52	29	1.33	23	1.58	21	1.65	30

Table III.2. Average bulk density and water contents of soil cores compacted at seven different target levels and maintained at seven water content levels in a 7x7 factorial arrangement.

Soil	Compaction Level	Average Bulk Density ($Mg\ m^{-3}$)	Water Level	Average Water Content ($cm^3\ cm^{-3}$)
Dome	1	1.04 ± 0.02	1	0.11 ± 0.03
	2	1.13 ± 0.01	2	0.13 ± 0.02
	3	1.26 ± 0.01	3	0.14 ± 0.02
	4	1.33 ± 0.01	4	0.18 ± 0.03
	5	1.42 ± 0.02	5	0.21 ± 0.04
	6	1.50 ± 0.01	6	0.27 ± 0.05
	7	1.54 ± 0.01	7	0.40 ± 0.09
Cohasset	1	0.81 ± 0.02	1	0.11 ± 0.00
	2	0.89 ± 0.02	2	0.17 ± 0.04
	3	0.96 ± 0.03	3	0.19 ± 0.04
	4	1.13 ± 0.00	4	0.24 ± 0.04
	5	1.19 ± 0.01	5	0.28 ± 0.05
	6	1.22 ± 0.01	6	0.34 ± 0.07
	7	1.26 ± 0.00	7	0.46 ± 0.11
Clarksville	1	1.13 ± 0.02	1	0.12 ± 0.02
	2	1.24 ± 0.02	2	0.14 ± 0.03
	3	1.29 ± 0.01	3	0.18 ± 0.04
	4	1.38 ± 0.01	4	0.23 ± 0.05
	5	1.45 ± 0.01	5	0.28 ± 0.06
	6	1.50 ± 0.01	6	0.34 ± 0.07
	7	1.53 ± 0.00	7	0.42 ± 0.12
Argent	1	1.27 ± 0.00	1	0.15 ± 0.04
	2	1.40 ± 0.01	2	0.22 ± 0.06
	3	1.48 ± 0.01	3	0.28 ± 0.08
	4	1.55 ± 0.01	4	0.33 ± 0.10
	5	1.61 ± 0.01	5	0.38 ± 0.13
	6	1.63 ± 0.00	6	0.44 ± 0.15
	7	1.65 ± 0.02	7	0.51 ± 0.18

The relationship between target ρ_b and actual ρ_b for all compacted soil columns was plotted on a 1:1 line (Fig. III.3). Soil column ρ_b values varied from the target ρ_b. Cohasset cores had lower than target ρ_b throughout its density range, Dome and Argent slightly higher than target densities and Argent soil columns were similar to target densities. The data fit reasonably well around the 1:1 line, suggesting this method of core compaction was adequate, given the large number of cores packed and subtle changes in water content during the process. Although the target to actual ρ_b variation was not the critical point of interest, this information does suggest this techniques efficacy for creating experimental cores at a range of targeted ρ_b.

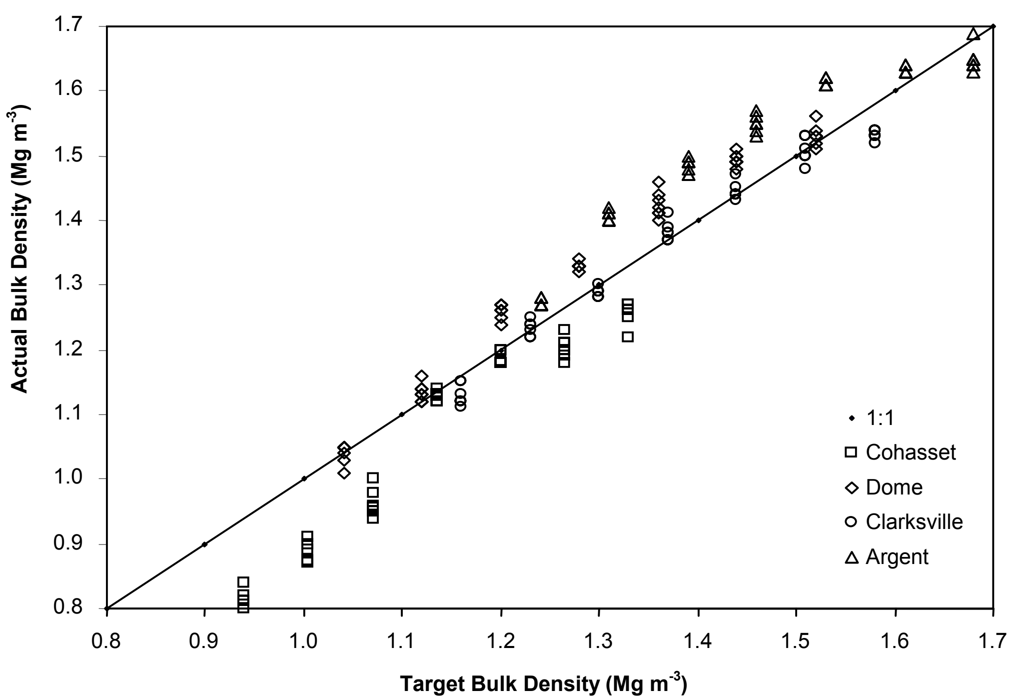

Figure III.3. Comparison of target to actual bulk density of compacted soil columns.

Soil Strength

Soil strength was measured in each soil column at the end of the experiment with a lab pocket penetrometer (BSE Model S-170). All cores were within their targeted volumetric water content range when measured. The core was placed on its side and the outer PVC core cut lengthwise in several places. The PVC segments were removed and triplicate soil strength measures taken and average soil strength for each pot determined. The flat-tipped pocket penetrometer with a standard 6.35 mm diameter tip was fitted with a smaller tip to measure the higher strength soils. Volumetric water content was determined for each soil column at the time of measurement. Soil strength as a function of ρ_b and θ_v was analyzed using multiple linear regression analyses.

Water and Aeration Characteristics

Soil Water Retention Curves and Soil Porosity

Pore size distribution and soil water retention curves were developed for all seven compaction levels within each soil type. Soil water contents at potentials of -0.005, -0.01, -0.03, -0.1 and -1.5 MPa were determined for each soil using standard tension table and plate techniques (Klute, 1986). PVC cylinders (8 by 10 cm) with compacted soil were used to determine Ψ_w for tensions up to -0.1 MPa, and 5 by 2.5 cm soil cores were used to determine water content at a Ψ_w of -1.5 MPa. Aeration porosity (0.00 to -0.01 MPa), available water (water between -0.01 and -1.5 MPa), and unavailable water (less than -1.5 MPa) were determined for all four soils along their density gradient.

Least Limiting Water Range

The least limiting water range, as used by da Silva and Kay (1997a), was developed for each soil using our experimental data. The upper LLWR limit is the lesser volumetric water content of field capacity (θ_{FC}) or aeration porosity < 10% (θ_{AP}), while the lower limit is the greater water content associated with either wilting point (θ_{WP}) or soil strength > 2.0 MPa (θ_{SS}) (Fig. II.2). Soil water retention curve data from each of the seven density levels were used to determine θ_{FC} and θ_{WP} critical limits. θ_{AP} was defined as total porosity minus 10%. The lower θ_{SS} limit was determined using Busscher's (1990) regression model as selected by da Silva and Kay (1994) describing the relationship of strength as a function of ρ_b and θ_v :

$$SS = c\ \theta_v^{\ d}\ \rho_b^{\ e} \qquad [Eq.1]$$

where θ_v is volumetric water content, ρ_b is bulk density and c, d and e are constants.

Soil Water Gradient

A θ_v gradient ranging from near-permanent wilting point to near-saturation was established for each soil. The weight of each pot associated with the target θ_v was calculated. Approximately every 3 days during the growth period (14 to 22 weeks), all pots were weighed and watered as necessary to maintain the target θ_v as closely as possible. If weight was below target, water was added to achieve the target θ_v; conversely, water was not added if the pot was too wet or within the range. Pot θ_v was maintained within a range of 10 to 15% of target θ_v with the exception of the wettest treatments for which the range was as large as 20 to 25% for some cores.

We also attempted to monitor volumetric water content with depth using time domain reflectometry (TDR) rods. Three 10 cm pieces of 0.31 cm ($1/8^{th}$) inch stainless steel welding rod were placed horizontally and permanently into the pot at 3 cm vertical intervals. Standard buriable waveguide TDR cables were adapted to connect with the metal rods. Although the technique appeared to have promise, we were not able to establish consistent relationships between the TDR moisture measures and actual θ_v; therefore, we did not use these data.

Seedling Establishment and Growth

Tree species normally found growing on each soil type were planted: Ponderosa pine (*Pinus ponderosa*) was grown on the Dome and Cohasset soils from California, shortleaf pine (*Pinus echinata*) on the Clarksville soil, and loblolly pine (*Pinus taeda*) on the Argent soil.

A 1 cm diameter hole was drilled in the center of each packed soil column to within 3.5 cm of the bottom. At planting time, this hole was back-filled with washed silica construction sand to allow rooting and water access to the soil column center along most of its depth. Fine mesh plastic screen was attached to the bottom of each core to prevent the loss of soil and allow water drainage. Columns were placed on a metal mesh greenhouse bench throughout the experiment. Greenhouse minimum and maximum temperatures and humidity were measured approximately once per week. The average weekly minimum temperature during the study period was 18 ^{o}C and the average maximum temperature was 33 ^{o}C. Humidity ranged from 50 to 93% with a weekly average of 56%.

Seed stock appropriate to the areas from which our soils were collected was used. The pine and oak seeds were planted in trays in a potting soil and sand mixture and were set in the greenhouse to germinate and grow. After 28 da, the most vigorous seedlings of approximately equal size were selected for planting. The seedlings were carefully planted in the center of each pot. Washed silica sand was added to the top of the soil to prevent soil surface disturbance from the watering treatments and to prevent the sand planting channel from clogging with soil. The seedlings were grown for six weeks with regular watering and nutrition to establish root growth prior to applying water stress. After the establishment period, seedlings were grown for the experimental period of approximately 13 weeks. We allowed the shortleaf pines on Clarksville to grow an additional 8 weeks because after 13 weeks these seedlings were still very small and roots had not reached the pot bottoms.

A commercial fertilizer (15-30-15) nutrient solution was foliar applied periodically in an attempt to provide adequate nutrition throughout the experiment. At each fertilizer application, each seedling received 4.5 ml of fertilizer solution containing 710 ppm N, 610 ppm P, 590 ppm K, 7 ppm Fe, 3 ppm Cu, 3 ppm Zn, 2 ppm Mn, 0.9 ppm B and 0.02ppm Mo. In addition, the pot bottoms were checked periodically for roots. Root pruning was observed at the bottom of several pots. Based on observations during harvest and root analyses, no root systems had fully exploited the soil in the pot and therefore were not soil resource limited. We also did not observe roots extensively using the zone between the soil and pot sides to grow. Roots moved out from the pots sand center into the soil matrix in almost all cores; however the extent of soil exploration was strongly related to treatment.

All seedlings survived the establishment phase. However, after water treatments were applied there was significant ponderosa pine seedling mortality on the Dome and Cohasset soils. Several shortleaf pines and many white oaks on the Clarksville soil died. No loblolly pines growing on the Argent soil died. We believe that the mortality was due to the watering treatment, particularly inadequate water for the ponderosa pines since seedlings grew well during the establishment period prior to application of the watering treatment.

Plant analyses

After the growing period, seedling height and root collar diameter were measured. Each core was then deconstructed and root systems separated from the soil by carefully washing with water. Root length and surface area were determined for each seedlings entire root system using a computer imaging analyzer (Delta T, Fisher, 1971). Root length density was determined. Shoot and roots were oven dried and both above- and below-ground biomass were measured and collected.

Model Development and Statistics

Multiple regression techniques were used to model root growth as a function of ρ_b and water content. We hypothesized that root growth would decrease linearly with increasing ρ_b (Foil and Ralston, 1967; Heilman, 1981; Mitchell et al., 1982). Root growth decreases from optimum both at the wet end and dry end of the soil water spectrum. Therefore, we hypothesized that this relationship could be depicted mathematically as a quadratic function. The basic model

$$\text{Root length density (RLD)} = b_0 + b_1 * \theta_v + b_2 * \rho_b + b_3 * (\theta_v)^2 \qquad \text{[Eq. 2.]}$$

was fit to each soil/species combination. We used regression analysis to also test if there was an interaction between ρ_b and θ_v. Terms were then added or deleted, based on their significance in the model, to reflect the observed data for each soil. Regression diagnostics (Cook's D and leverage analysis) were used to examine the influence of outliers on the model shape. Plots of the residuals were evaluated to assess model fit. Simple linear regression was used to test shoot growth as a function of root growth for each soil-species combination. Mean RLD of ponderosa pines growing on Dome and Cohasset soils were compared with a t-test. Seedling growth in and out of the LLWR was compared with a t-test. Analysis of variance was used to test differences in growth parameter means for the seven compaction levels. All statistical analyses were performed using the SAS statistical software (SAS Institute Inc., Cary, N.C.).