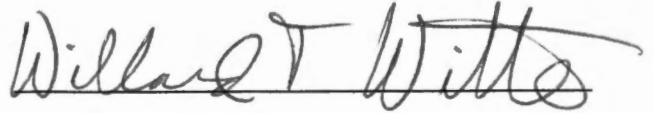


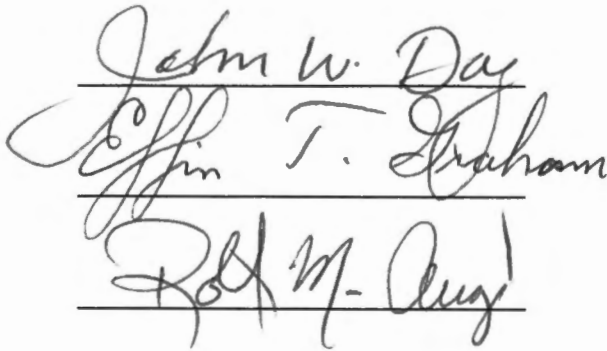
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Willard T. Witte, Major Professor

We have read this thesis
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**ROOT REGENERATION OF CHEMICALLY
ROOT PRUNED WOODY ORNAMENTALS**

A Thesis

Presented for the

Master of Science

Degree

The University of Tennessee, Knoxville

Phillip Clay Flanagan

August 1991

AD-VET-MED.

THESIS
91
.F572

To my wife Rosilyn,

for her love and friendship.

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ABSTRACT

These experiments were designed to study the effects of alternative copper compounds in a latex paint carrier as root pruning agents on several container-grown woody ornamental plants and subsequent physiological effects on root regeneration.

Quercus acutissima, (sawtooth oak) seedlings were used to study chemical root pruning effects and subsequent root regeneration. Interior surfaces of tube trays were painted with exterior acrylic latex paint (LP) containing 0, 47, 90 or 228 g/l tribasic copper sulfate (TCS). After 15 weeks, only 0.7 roots per seedling continued growth after being deflected by the tubewall of the 90 g/l TCS treatment compared with 3.7 for the control. Fibrous roots on plug surfaces were reduced when in contact with TCS treated container walls. Height and caliper were not affected at any TCS level. Three weeks after transplanting to larger untreated containers, height and caliper were still unaffected by any TCS treatment. Time required for regeneration of new roots was not affected by TCS treatments.

Rooted, bed-grown cuttings of Viburnum tomentosum plicatum 'Mariesii' (viburnum) were used to evaluate the effects of copper hydroxide (CH) on root pruning and root regeneration. Interior walls of 3 l containers were painted with LP containing 0, 47, 90, 185 and 260 g/l CH. Plants were grown 5 months in a pine bark medium in these containers or control (0 LP, 0 CH) containers. Fewer roots continued growth after contact with container walls in the CH treatments compared with the control treatment. There were 61, 15, 12, and 9 roots/plant in the 47, 90, 185, and 260 g/l CH treatments respectively, while 112 roots/plant continued to grow in control treatments. Elemental

analysis of serial root sections showed Cu concentrations highest in root tips with concentrations rapidly decreasing proximally. After transplanting from treated containers, 100% root regeneration of selected roots occurred within 2 days in control plants; 88% in 4 days for LP only; 92% in 4 days for 47 g/l CH; 98% in 4 days for 90 g/l CH; 76% in 4 days for 185 g/l CH; and 92% in 6 days for 260 g/l CH. After 32 days plants in the 90 g/l CH treatment reached the same level of root growth as control plants. Statistical root growth curves were established for each CH treatment. Micrographs of medial longitudinal sections of root tips in the 90 g/l CH treatment showed cells were becoming disorganized in the apical meristem. Cells within the vascular cylinder and cortex were maturing quicker than those in the control treatments. At 260 g/l CH, root cap and apical meristem cells were replaced with a callus-like mass of non-organized cells. Root tips were thickened and club-like from contact with CH treated container walls.

Rooted cuttings of Euonymus alatus 'Compacta' were grown 6 months in 0.9 l containers painted with CH to determine long term effects on secondary branching of roots in contact with treated surfaces. Harvest height of the shoot portion of the plant was not affected at any CH level. Mean distance between roots decreased in copper treated plants as CH levels increased. Fresh weights (FW) and dry weights (DW) were 12% and 18% (shoots and roots) heavier in the 90 g/l CH treatment than in control plants.

TABLE OF CONTENTS

CHAPTER I	1
INTRODUCTION	1
CHAPTER II	3
REVIEW OF LITERATURE	3
Root Distribution	3
Root Pruning	4
Root Regeneration	5
Container Production	6
Grow-Bag Production	8
Chemical Root Pruning	9
Elemental Copper	12
CHAPTER III	14
EFFECTS OF $\text{Cu}_4\text{H}_6\text{O}_{10}\text{S}$ AS A CHEMICAL ROOT PRUNING AGENT	14
Introduction	14
Materials and Methods	14
Results and Discussion	22
CHAPTER IV	26
EFFECTS OF $\text{Cu}(\text{OH})_2$ AS A CHEMICAL ROOT PRUNING AGENT	26
Introduction	26
Materials and Methods	27
Results and Discussion	34

CHAPTER V	50
EFFECTS OF $\text{Cu}(\text{OH})_2$ ON SECONDARY BRANCHING OF ROOTS	50
Introduction	50
Materials and Methods	50
Results and Discussion	52
CHAPTER VI	58
CONCLUSIONS AND SUMMARY	58
General Conclusions	58
Summary	60
LIST OF REFERENCES	61
VITA	69

LIST OF TABLES

TABLE

1.	Concentration and dilution schedule for incorporating tribasic copper sulfate (TCS) in water prior to mixing with exterior acrylic latex paint (LP).	17
2.	Mean number deflected roots per plug and mean rating of fibrous roots on plug surface of <u>Quercus acutissima</u> (sawtooth oak) seedlings grown 15 weeks in plug trays painted with tribasic copper sulfate (TCS)/exterior acrylic latex paint (LP) on interior walls	23
3.	Mean shoot height and number of lateral roots regenerating from tap root of <u>Quercus acutissima</u> (sawtooth oak) seedlings grown 15 weeks in tribasic copper sulfate (TCS)/exterior acrylic latex paint (LP) treated trays and 3 weeks in untreated 3 l containers.	24
4.	Concentration and dilution schedule for incorporating cupric hydroxide (CH) in water prior to mixing with exterior acrylic latex paint (LP).	29
5.	Mean rating of root growth on root ball surface of <u>Viburnum tomentosum plicatum</u> 'Mariesii' grown 5 months in 3 l containers painted with cupric hydroxide (CH)/exterior acrylic latex paint (LP) on interior walls. . .	35
6.	Mean number of deflected and chemically root pruned roots of <u>Viburnum tomentosum plicatum</u> 'Mariesii' grown 5 months in containers painted with cupric hydroxide (CH)/exterior acrylic latex paint (LP) on the inside walls.	36
7.	Mean rating of root growth on root ball surface of <u>Viburnum tomentosum plicatum</u> 'Mariesii' grown 5 months in 3 l cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers, and 8 months in untreated 12 l containers.	38
8.	Statistical root growth curve parameters established for slope (B_1), exponential growth (X_1) and asymptote (A) for <u>Viburnum tomentosum plicatum</u> 'Mariesii' grown in cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers for 5 months and untreated plexiglass growth trays for 32 days.	41

TABLE

9.	Concentration analyses (ppm) of selected elements in root tissues of <u>Viburnum tomentosum plicatum</u> 'Mariesii' grown in cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers 5 months (+) and after 32 days in untreated containers (-).	43
10.	Comparison of mean number of lateral branching roots within the first 2.5 cm of the root tip, and mean mm distance between first 5 branch roots of <u>Euonymus alatus</u> 'Compacta' grown 6 months in cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers.	53
11.	Grams fresh weight (FW) and dry weight (DW) of <u>Euonymus alatus</u> 'Compacta' shoots and roots of plants produced in cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers for 6 months. . . .	54

LIST OF FIGURES

FIGURE

1.	Sawtooth oak acorns on pine bark medium.	16
2.	#38 tree trays painted with TCS/LP treatments on interior tube walls .	18
3.	A sawtooth oak seedling grown 15 weeks in #38 tree trays showing a deflected root which continued to grow after contacting TCS/LP treated walls.	20
4.	Bed-grown, rooted cuttings of euonymus (left photograph) and viburnum (right photograph).	28
5.	Root regeneration growth curve of viburnum, chemically root pruned with CH/LP treatments, after 32 days in untreated plexiglass growth trays.	39
6.	Microphotographs of medial, longitudinal root tip sections of viburnum.	49
7.	Cleaned euonymus root balls, suspended in water, grown 6 months in CH/LP treated containers.	56
8.	Multiple branching behind root tip of euonymus in contact with 260 g CH/LP after 6 months in treated containers.	57

CHAPTER I

INTRODUCTION

The toxic physiological effects of copper on root growth have long been recognized. For instance, the municipality of Ridgewood, New Jersey used copper sulfate in 1937 to eliminate roots from sewer lines (27). Twice each year, 1 liter of 6 mm crystals was flushed through porcelain toilet bowls in each residence into sewer lines infested with tree roots. Roots were eliminated without any apparent injury to the trees being treated.

Forestry research using metal compounds as chemical root pruning agents began in the mid-1960's (50) and continued through the 1970's (6, 11, 22). In the early 1980's research by Burdett *et al.* (11, 12, 13), McDonald *et al.* (44, 45, 46), Romero (57), and Ruehle (58) used copper carbonate in combination with an exterior latex paint (LP) carrier as a chemical root pruning agent in plug trays for seedling conifer production in the Pacific Northwest.

Several forms of metal compounds have been studied for effectiveness as chemical root pruning agents. Pellet *et al.* (51) studied more than 15 metal compounds, testing their effectiveness for chemical root pruning. Effectiveness was dependent on chemical compound, species, method of application, concentration levels and type of media. Recently horticulturists have experimented with copper compounds mixed with a carrier, usually LP, as a chemical root pruning paint in larger containers (3, 4, 5, 68, 78, 79).

The goal here is to find the best overall chemical root pruning agent for production of woody ornamental plants grown in large scale systems. Root system development and physiology has an impact on the landscape industry's ability to plant woody ornamentals on a commercial basis year-around. Through chemical root pruning research it may be possible to grow and install plant materials successfully throughout the year. The key to this will be the development of a plant root system that, when placed in the landscape, will provide stable anchorage for future plant development and provide rapid root regeneration to help insure survival and growth. Researchers must be aware of growth and physiological factors in woody ornamental plants, including, but not limited to: root distribution, root pruning, root regeneration, and various types of container production systems.

Cost of materials must be considered. CuCO_3 is expensive, about 10 times the cost of $\text{Cu}_4\text{H}_6\text{O}_{10}\text{S}$, (\$60.00/lb vs \$6.00/lb), and is often difficult to obtain through ordinary channels.

This research project was designed to study the effects of alternative copper compounds in a LP carrier as root pruning agents. Several container-grown woody ornamental plants were used in order to observe any variation in root regeneration, plant growth and root cellular structure. Two forms of copper, $\text{Cu}_4\text{H}_6\text{O}_{10}\text{S}$ (tribasic copper sulfate [TCS]), and $\text{Cu}(\text{OH})_2$ (cupric hydroxide [CH]), were used in conjunction with LP as a carrier for the copper, with concentrations ranging from 47 to 260 g/l of LP.

CHAPTER II

REVIEW OF LITERATURE

Root Distribution

The major functions of tree and shrub root systems are water and mineral absorption, anchorage and storage (43). Woody plants have two basic types of root systems; tap roots and fibrous roots (35). Development of tap roots or fibrous roots are dependent on tree species, soil condition and type, water availability, fertility, temperature and cultural practices (27). Optimum conditions for root growth vary among woody species and are related to available soil moisture and aeration.

According to Perry (53), "...approximately 99% of the roots occur within the surface meter of the soil and extend outward over an area one to two or more times the height of the tree." During harvesting of field grown nursery stock 80 to 90% of the plants' root systems may be left in the soil outside of the root ball (26, 53, 76). Watson and Himelick (76) investigated 6 species of trees and found that a typical tree (4 m high and 10 cm caliper) had a root system which utilized about 20 m³ of soil for water and mineral absorption. Upon transplanting, soil volume of the root ball was reduced to 0.35 m³. When a tree is dug, the roots remaining within the root ball are older suberized roots. These roots are able to absorb water and minerals, but the volume of soil from which they absorb water has been drastically reduced (1, 34, 74). Rapid root regeneration after transplanting increases the plants' ability to obtain water and minerals in soil (37).

The most serious factor modifying root distribution of woody ornamental plants, especially trees, is soil compaction (53). Construction equipment is a major contributor to this problem but so are large farm animals, heavy foot traffic and parking of vehicles in non-paved areas. The compaction process reduces pore space essential for absorption of air and water (53). Oxygen and water may also be restricted in soils by the use of plastic mulches.

Root Pruning

Root pruning is unavoidable in harvesting field grown nursery stock. An immediate reaction to root pruning is a reduction in uptake of water and minerals due to loss of absorptive surface area (23). Restricted water uptake may result in a water deficit if transpiration exceeds water absorption. Sustained water deficits usually result in stomatal closure and reductions in transpiration (35). Temporary declines in photosynthesis have been observed in pine trees after root pruning, but recovery was noted within 4 weeks (23). Root pruning was effective in promoting dense, compact root systems (26), stimulation of flowering and fruiting, acclimation to drought stress and dwarfing of some plants (23).

Root:shoot ratios are genetically and environmentally determined (35, 43). They remain constant under natural conditions and decrease progressively with age and size of tree. Field grown nursery stock which has not been root pruned must regenerate its root system after the plant is set in the landscape. For large trees, reestablishment is inhibited by severe root loss during field harvest (75), creating an imbalance between roots and

shoots. The more roots lost, the greater the imbalance and the slower the reestablishment process (75). As plants attempt to restore root:shoot ratios, assimilates are transported to the root system to increase root growth while shoot portions of the plant experiences reduced growth (23). In an attempt to compensate for losses in root systems, leaves and branches may be shed to reestablish root:shoot balance (52).

Root Regeneration

Rapid root regeneration in woody ornamentals after transplanting is a key factor in plant survival (23, 39, 66, 76, 77, 82). Rapid root regeneration shortens the duration and severity of growth interruption (43).

Several factors influence root regeneration and establishment of plants in the landscape. Differences among species relate to problems with root regeneration capacity and/or root system viability at certain times of the year. Root growth rates vary within a species in any given year due to site, environment, and location of roots within the plants own root system (35). Woody plant root growth is basically driven by the energy resources of the plant, influenced by temperature and moisture availability. Root growth peaks during spring to early summer and again in fall, with most new root regeneration occurring in spring (23, 43, 56). Root growth in the fall consists primarily of elongation of existing roots (63). Therefore, species such as Quercus coccinea (scarlet oak) and Liriodendron tulipifera (tulip poplar) , which have coarse root systems, transplant easier in the spring when there is both initiation of new roots and elongation of existing roots (32, 66).

Another factor affecting plant reestablishment is size of the propagation container (2). Cedrus deodara (Deodar cedar), Pinus taeda (loblolly pine) and Pinus thunbergiana (Japanese black pine) seeds propagated in 0.68 l containers and transplanted into 3.8 l white poly bags, grown for 14 months, had significant increases in height, caliper and number of branches compared to seeds propagated in 0.36 l containers and transplanted into 3.8 l containers and grown for the same period. Seasonal effects on root regeneration have been reported on Pinus nigra (Austrian pine) and Thuja occidentalis (arborvitae) (38). Poor root regeneration and high needle water deficits occurred in fall transplantings of Austrian pine. Spring transplantings, after chilling requirements for dormancy release were met, provided best root regeneration. Arborvitae transplanted in fall had good root regeneration, but spring transplants had better root regeneration. Lathrop and Mecklenburg (40) determined that root regeneration of Taxus cuspidata was dependent on separate chilling requirements for root dormancy and bud dormancy. Adequate carbohydrates and the presence of non-dormant buds to provide auxins, or other chemicals, may be required for stimulation of cambial activity which in turn stimulates lateral root primordia, as has been demonstrated for Pistacia chinensis (41).

Container Production

Even though container grown plant materials retain their entire root system, problems still arise with root-bound plants or distorted roots which may be kinked or coiled (17, 28, 65). According to Davidson *et al.* (17), deformed roots are more susceptible to drought. Deflected roots in container grown pines are not able to provide

anchorage for the tree after planting out, leading to basal sweep and wind throw as the tree matures (48). Conifers grown in small containers and tubes are more prone to death, subject to trunk breakage and wind throw than trees naturally established (28, 48). Root pruning is typically used to correct root system deformities of container plants (5). However, root pruning may lead to other problems such as reduced photosynthesis (24), reduced transplant survival (39), and reduction in plant growth for three or more years (18, 23). In container production systems root:shoot relationship is a major factor in the failure of many trees and shrubs set in the landscape (27). Heavy fertilization and watering of container stock produces plants with a large leaf area in relation to root mass in containers. Water retention within the root ball is reduced after transplanting , but foliage continues to transpire causing water deficits (27).

Whitcomb (80, 81), developed several styles of specialized containers to eliminate problems of root kinking and coiling associated with container production. His early container designs were developed with vertical slits in the sides to air root prune circling roots and promote secondary branching, which would increase root surface area for absorption of water and nutrients. RootMaker™ and RootBuilder™ are two brands of containers based on air root pruning. Tap roots of seedlings grown in RootMaker™ containers (6 cm square X 10 cm deep) are directed toward holes and air root pruned, which stimulates formation of lateral roots on the main tap root and, in turn, secondary lateral roots are air root pruned along the stair-stepped sides of the container. RootBuilder™ containers are modular panels (26 l to 643 l) with 200 hexagonal shaped funnels in the sidewalls of each panel. Air root pruning takes place at the container wall

container wall and secondary branching occurs within the media.

Grow-Bag Production

Grow-bags are a modification of field culture. Trees are grown in soil and the root system is confined by a non-woven fabric grow-bag. Some roots penetrate the fabric but are girdled. Girdled roots tend to generate secondary roots within the grow-bag. Recent research indicated this method can produce plants that suffer less shock at transplant, but there are certain limitations (21, 31, 69, 83). Ingram *et al.* (31), reported that effectiveness of grow-bags is species dependent. They found no growth advantages for Ulmus parvifolia (Drake elm), Lagerstoemia indica (crape myrtle), or Pinus elliottii (slash pine) in grow-bags, but Liquidambar styraciflua (sweetgum) and Quercus virginiana (live oak) responded favorably under their experimental conditions. Early grow-bag models had a plastic bottom sewn to a non-woven cloth sidewall (15, 55), but roots penetrated through the stitching and became dominant outside the grow-bag. Rapidly growing trees in grow-bags may develop roots circling within the bag (70, 71). Acer saccharinum (silver maple) and Liriodendron tulipifera (tulip poplar) whips grown in 14 inch grow-bags for 2 years had less height and caliper compared to those in conventional field production, while Cornus florida (dogwood) and Platanus occidentalis (sycamore) did not. Fast growing trees may suffer growth losses if held in 14 inch grow-bags longer than 2 years (71). Changes in bag design and proper choice of bag size for the type of tree and length of production period may reduce these problems (54).

Chemical Root Pruning

The use of copper compounds for chemical root pruning container grown plants has had varying degrees of success. While excess copper is toxic to plants, it is important to remember that tolerance to copper and other heavy metals varies within and among species (14).

Early research showed the effect of copper carbonate on mycorrhizal fungi and pine seedling development. Root tips coming in contact with copper treated surfaces stimulated lateral roots resulting in greater root surface area. Subsequent mycorrhizal inoculation of the increased surface area further enhanced absorption capacity (46, 58). Mycorrhizal fungi aid plants by increasing nutrient uptake, including copper, in nutrient poor or nutrient-fixing soils (33).

Other forms of copper have been used for root control. Saul (61) used sheets of metallic copper, copper-armored fiber and copper paint to control roots of tube-grown seedlings. Copper naphthenate and copper sulfate controlled roots without phytotoxicity in Jacaranda acutifolia (jacaranda) and Eucalyptus viminalis (eucalyptus) (22).

The lack of mechanical stability in plug grown pine seedlings led Burdett (11, 12) to study the effects of chemical root pruning on container grown Pinus contorta (lodgepole pine). A mixture of copper carbonate and LP was painted on the inside surface of styroblock containers. Treated container walls effectively inhibited elongation of lateral roots and eliminated the tendency for roots to grow downward and circle. When treated seedlings were planted in the field, root regeneration occurred rapidly and root system conformation was comparable to seedlings of naturally established trees. Top growth of

treated plants remained similar to untreated plants for the first two years. In the third and fourth year, top growth of treated seedlings was 15% greater than untreated plants (12).

In a separate experiment by Burdett *et al.* (13), seedlings of several conifer species were treated with the same copper carbonate/LP mixture. In most cases, chemically root pruned plants showed vigorous root growth after transplanting. Effectiveness of chemical root pruning treatments varied with species, container size, growing medium and copper concentrations. McDonald *et al.* (44, 45) substantiated Burdett's results when they duplicated the experiments.

Arnold and Struve (4, 5) produced container-grown, landscape size trees of Fraxinus pennsylvanica (green ash) and Quercus rubra (red oak), utilizing copper carbonate as a chemical root pruning agent. Treated green ash seedlings in small containers had a higher root regeneration potential than controls (4). Copper concentrations within the plant varied, with highest levels occurring in root tips and lowest levels in foliage. Method of treatment was by mixing copper carbonate with LP and applying to inner container walls.

In a second experiment (5), root regeneration and shoot growth following transplanting of copper-root-pruned green ash and red oak from 2.2 l into 45.6 l plastic bags was evaluated. Mechanical root pruning was not required on plants grown in 2.2 l containers treated with copper carbonate. Plants grown in untreated containers lost up to 37% (DW) of their root system due to root pruning required to correct root deformations caused by circling of roots within containers. After plants were transplanted to 44 l containers, those treated with copper carbonate had more DW of regenerated roots

beyond the original container boundary compared to untreated plants. After two years in the field, plants from copper carbonate treated containers showed enhanced shoot growth compared with those root pruned.

Pellet *et al.* tested various other forms of metal compounds as chemical root pruning agents (51). Chemical compounds were incorporated with vermiculite and then a thin layer of the mixture placed at various depths within media to test chemical root pruning properties. Cobalt chloride, cupric sulfate, nickel chloride, silver nitrate, sodium borate and zinc sulfate were used to successfully root prune Cucurbita pepo (summer squash), Phaseolus limensis (lima beans) and Gymnocladus dioica (Kentucky coffeetree). Field studies were conducted on Cercis canadensis (redbud) and Catalpa bignoniodes (catalpa) seedlings by applying silver nitrate solutions on soil surfaces and placing a frame around the treated area prior to filling with soil. Both species were effectively root pruned. Plant species, chemical concentrations, type of media and length of growing time were factors influencing this study. Ineffective metal compounds were: aluminum sulfate, barium sulfate, lithium chloride, magnesium chloride, magnesium sulfate, manganese chloride, manganese sulfate, potassium permanganate and stannous chloride.

Elemental Copper

Copper is a micronutrient normally found in soils in the divalent form and absorbed by plants in the divalent form or as salts from organic complexes (47,72). The average content of copper in soil varies from 2 ppm to 150 ppm (10, 72). From a biochemical standpoint, copper acts as a metal activator of several enzymes such as

tyrosinase, laccase and ascorbic acid oxidase, and serves as a catalyst for respiration, chlorophyll synthesis, carbohydrate and protein metabolism (47, 59, 72).

In soils, copper is strongly bound to colloids and not readily mobile. As organic matter increases in soils so does retention of copper (10, 47, 72). A soil pH above 7 is considered a critical point for reduction of copper availability (10). Over-liming reduces availability of copper, but also iron, manganese and zinc.

Normal copper concentrations in plants are 2 to 20 ppm in air or oven dried plant material (47). Deficiency symptoms vary in plants. In cereal crops, first signs are noticed in leaf tips when they become white and leaves are narrow and twisted. In some trees, a weeping or pendulous form may be exhibited due to the lack of copper which is required in the synthesis of lignin, causing inadequate development of the xylem (47).

Toxic levels of copper, and other heavy metals in plants are variable within and among species (14). At toxic levels, copper is capable of replacing other metal ions, especially iron, giving plants a chlorotic appearance (16). Copper has the ability to displace most other ions from root exchange sites and become strongly bound in root free spaces yielding higher copper concentration levels within roots of plants (29). The most rapid response to toxic levels of copper is inhibition of root growth (47). By measuring K^+ leakage in root cells, it was determined that excess copper has detrimental effects on the plasmalemma of roots (73).

Copper compounds differ in molecular weight and solubility. Cupric carbonate (Malachite), has a molecular weight of 239, cupric hydroxide, a molecular weight of 98 and tribasic copper sulfate a molecular weight of 453. All are insoluble in water, but are soluble in the presence of dilute acids and ammonia (63).

CHAPTER III

EFFECTS OF $\text{Cu}_4\text{H}_6\text{O}_{10}\text{S}$ AS A CHEMICAL ROOT PRUNING AGENT

Introduction

Copper carbonate has been used in most of the past research on chemical root pruning. However, the relatively high cost of that product may limit its practical use. Cost fluctuates from time to time, but it is normally 8 to 10 times more expensive than tribasic copper sulfate (TCS).

This experiment was designed to test the effects of TCS mixed with LP as a chemical root pruning agent on the inside wall of containers. Seedlings grown in containers painted with TCS mixtures were evaluated for the ability of TCS to 1) reduce or eliminate the number of roots continuing to grow after contacting treated tube walls, 2) influence height and/or caliper along with fresh weight (FW) and dry weight (DW) of shoots and roots of seedlings during chemical root pruning or after a period of regeneration in untreated containers, 3) increase secondary and lateral root growth behind root tips in contact with treated surfaces.

Materials and Methods

This experiment was arranged in a randomized complete block design with 5 treatments and 6 replications with 5 seedlings per treatment.

Quercus acutissima, sawtooth oak, acorns were collected on September 16, 1989 and placed in flats filled with pine bark medium. Acorns were covered with black fabric weed barrier to retain moisture and maintain even temperature, placed on raised benches in a double-layer, poly-covered greenhouse and watered as needed to prevent media and acorns from drying out (Figure 1). First germination occurred the week of November 29, 1989. On February 1, 1990, TCS¹ was incorporated with LP². TCS was incorporated in tap water to facilitate mixing (Table 1). Each TCS mixture was painted onto the interior walls of #38 Tree Trays³, and allowed to air dry (Figure 2).

One hundred fifty seedlings were selected for uniformity with shoot length 10 to 12 cm and root length 8 to 10 cm. On February 2, 1990, seedlings were divided into 6 replications with 5 seedlings in each of the 5 treatments and planted in ground pine bark media. Each m³ of pine bark was amended with: 4.17 Kg dolomitic limestone, 1.19 Kg treble superphosphate, 1.19 Kg 10-10-10 granular fertilizer, 1.34 Kg gypsum (CaSO₄), 0.89 Kg Micromax⁴ and 1.19 Kg epsom salts (MgSO₄). No additional fertilizers were used during the chemical root pruning portion of the experiment.

¹ Tennessee Chemical Company, Copperhill, TN

² Glidden Spread House - 100% Acrylic Latex No. 3600 White:
Glidden Coatings and Resins, Cleveland, OH

³ Tree Masters of Florida, Sydney, FL

⁴ Grace Sierra, Milpitas, CA

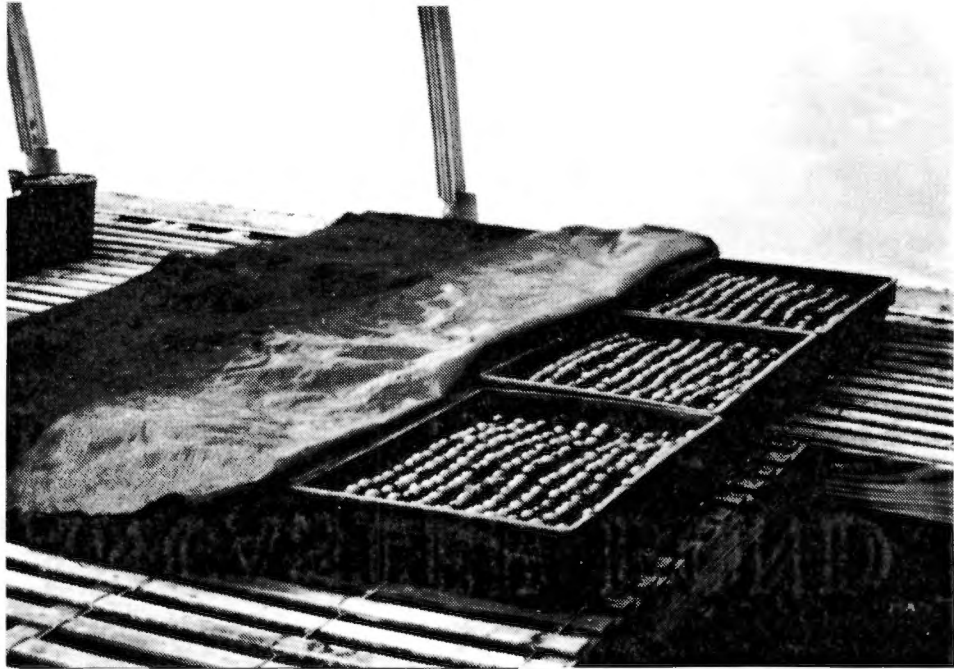


Figure 1. Sawtooth oak acorns on pine bark medium. Black weed barrier was used to maintain moisture and temperature.

Table 1. Concentration and dilution schedule for incorporating tribasic copper sulfate (TCS) in water prior to mixing with exterior acrylic latex paint (LP).

Treatment	Grams TCS	Water (l)	LP (l)	Final Volume (l)	Grams TCS/l LP (w:v)
1	0	0	0	0	0
2	0	0	1.0	1.0	0
3	50	0.05	1.0	1.06	47
4	100	0.08	1.0	1.10	90
5	300	0.20	1.0	1.24	228

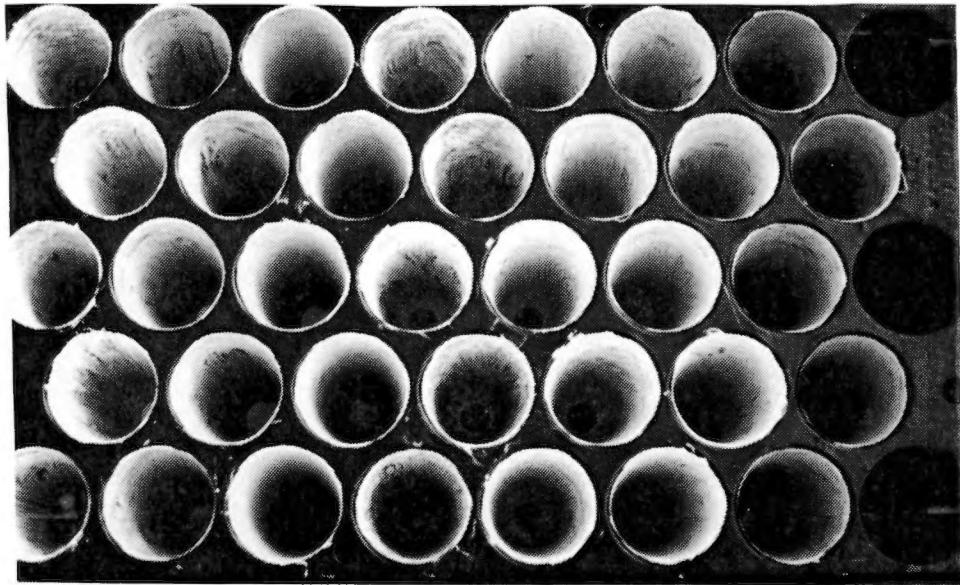


Figure 2. #38 tree trays painted with TCS/LP treatments on interior tube walls.

Seedlings were grown in a heated fiberglass greenhouse for 15 weeks. Average day/night temperatures were $22.2^{\circ}\text{C}/12.2^{\circ}\text{C} \pm 1.7^{\circ}\text{C}$. To grow the seedlings under 18 hour days, a 400W high pressure sodium lamp⁵ was placed 106 cm above the top of the tree trays.

On May 17, 1990, height and caliper of all seedlings was recorded. Root plugs were removed and the number of roots deflected and continuing to grow after contacting tube walls were counted (Figure 3). Fibrous roots on the outside of each plug were assigned a rating of 1 for minimal fibrous roots, 2 for moderate and 3 for heavy fibrous roots.

One seedling from each replication and treatment (30 total) was selected and stems and roots weighed to the nearest 0.1 gram on a Model 1200P balance⁶. Stems were severed at the top of the tube tray. Media was washed from each root plug and roots gently blotted dry with a towel before FW was recorded. Stems and roots were bagged and placed in Stabil-therm drying oven⁷ at 105°C for 48 hours.

Single seedlings were selected from each replication and treatment (30 total) and transplanted into the field research nursery on May 18, 1991. Data will be recorded at the end of the 1991 growing season and analyzed to determine effects of TCS treatments on plant growth and root regeneration. These trees were fertilized with 3 g Nursery

⁵ Voight Lighting Industries, Philadelphia, PA

⁶ American Scientific Products, McGaw Park, IL

⁷ Blue M Electric, Blue Island, IL



Figure 3. A sawtooth oak seedling grown 15 weeks in #38 tree trays showing a deflected root which continued to grow after contacting TCS/LP treated walls.

Special 12-6-6⁸ per tree at 8 to 10 week intervals during the growing season. Supplemental watering was by overhead irrigation.

The 3 remaining seedling groups (30 seedlings per group) were transplanted into untreated, 3 l black plastic containers⁹ and placed in the container research nursery area. Seedlings were fertilized at 5 day intervals with Peter's 20-20-20¹⁰ at the rate of 200 ppm N. Supplemental watering was done by hand.

At weekly intervals 1 group of seedlings (30 total) was removed and data recorded for height, caliper and FW and DW of stems and roots. Height was measured from the media surface to the apical bud. Caliper was measured 2.5 cm above the media surface with a Digimatic caliper¹¹. Stems were severed at the media surface and tree plugs were cleaned as previously described. The number of roots regenerated from the first 3 cm of the tap root were counted. Shoots and roots were dried by the same process previously described.

Data were analyzed using Statistical Analysis Software¹² Means were separated by Waller-Duncan K-ratio T test at the 5% significance level.

⁸ StaGreen, Sylacauga, AL

⁹ Zarn 300X, Zarn Inc., Reidsville, NC

¹⁰ W.R. Grace and Co., Fogelsville, PA

¹¹ Mitutoyo, Japan

¹² SAS 6.03, SAS Institute, Inc., Cary, NC

Results and Discussion

There were no differences in height, caliper, or FW and DW of roots due to any treatment level at termination of chemical root pruning.

Table 2 shows all levels of TCS controlled root deflection. In the most effective treatment, only 0.7 roots per seedling continued to grow after being deflected by tube walls painted with 90 g/l TCS, compared with 3.7 roots and 2.4 roots in control and LP only treatments. The increase in deflected roots in the 228 g/l treatment, as compared to the 90 g/l TCS treatment cannot be explained.

Surface fibrous roots on the plug were reduced by contact with copper treated container walls (Table 2).

Table 3 shows the tallest plants after 3 weeks growth following transplanting to untreated containers were from the lowest level of TCS treatment, 47 g/l LP only, 90 and 228 g/l TCS were the shortest. Control treatments were intermediate. There were no differences in harvest caliper at any treatment level 3 weeks after transplanting from plug trays.

The lowest level of TCS caused more lateral root regeneration in the first 3 cm of the tap root than the highest level (Table 3). After cleaning of media from the root ball, visual inspection showed increased lateral and secondary root growth on roots inhibited by contact with treated tube walls. Configuration of the root system was better in seedlings treated with TCS. Increased lateral and secondary root growth should reduce stress when plug seedlings are planted in the field by providing more root surface area

Table 2. Mean number deflected roots per plug and mean rating of fibrous roots on plug surface of *Quercus acutissima* (sawtooth oak) seedlings grown 15 weeks in plug trays painted with tribasic copper sulfate (TCS)/exterior acrylic latex paint (LP) on interior walls.

Treatment	Grams TCS/LP	# deflected roots per plug ^z	Rating of fibrous roots ^z
1 Control	0	3.7 a	2.8 a
2 LP Only	0	2.4 b	2.5 b
3 TCS + LP	47	1.4 c	1.3 c
4 TCS + LP	90	0.7 d	1.1 c
5 TCS + LP	228	1.4 c	1.2 c

GLM Procedure, Waller-Duncan K-ratio T test.

^z Means within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

Table 3. Mean shoot height and number of lateral roots regenerating from tap root of *Quercus acutissima* (sawtooth oak) seedlings grown 15 weeks in tribasic copper sulfate (TCS)/exterior acrylic latex paint (LP) treated trays and 3 weeks in untreated 3 l containers.

Treatment	Grams TCS/LP	Height ^z (cm)	# of lateral roots ^z
1 Control	0	24.7 bc	4.2 ab
2 LP Only	0	23.0 cd	4.2 ab
3 TCS + LP	47	29.3 a	6.6 a
4 TCS + LP	90	23.3 cd	3.9 ab
5 TCS + LP	228	21.7 d	3.2 b

ANOVA Procedure, Waller-Duncan K-ratio T Test.

^z Means within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

for absorption of water and minerals. Stimulation of root growth should provide a root system capable of stabilizing the tree in future years.

CHAPTER IV

EFFECTS OF $\text{Cu}(\text{OH})_2$ AS A CHEMICAL ROOT PRUNING AGENT

Introduction

Past researchers have used copper carbonate (CuCO_3), concentrations from 25 to 500 g/l of LP, to chemically root prune conifer seedlings (11, 12, 13, 45, 58). Effectiveness of root pruning varied with species, container size, growing media and concentration (45, 52). Rapid root regeneration after removal from treated containers was observed along with development of a root system comparable to seedlings of naturally established trees (12). However, copper carbonate is relatively expensive and not readily available through normal supply channels. Cupric hydroxide, $\text{Cu}(\text{OH})_2$, (CH), is a low cost and readily available fungicide.

The objectives of this study were to evaluate effectiveness of various concentrations of CH in a LP carrier as a chemical root pruning agent for container grown woody ornamental plants. Additional objectives were to 1) measure rate of root regeneration of chemically root pruned plants, 2) evaluate effects the of CH on other nutrient elements within selected plant tissues and 3) to observe effects of CH on internal cell structure of root tips.

Materials and Methods

This experiment was a randomized complete block with 6 treatments, 20 replications and 2 plant species.

Dormant rooted cuttings of Viburnum tomentosum plicatum 'Mariesii' (viburnum) and Euonymus alatus 'Compacta' (euonymus) were received from Schaefer Nursery, Winchester, Tennessee in mid-February 1990 and placed in cold storage at 4 °C. On March 29, 1990, 120 dormant rooted cuttings of each species were selected for uniformity. Roots were pruned to 8 cm and shoots to 8 cm (Figure 4).

On March 30, 1990 technical grade CH¹³ was mixed into LP¹⁴. CH was first mixed in tap water to facilitate mixing with LP (Table 4). Each CH treatment was painted onto interior walls of 40, 3 l, black plastic containers¹⁵ and allowed to air dry.

Ground pine bark was used as the planting medium. Each m³ of pine bark was amended with: 4.17 Kg dolomitic limestone, 1.19 Kg treble superphosphate, 1.19 Kg 10-10-10 granular fertilizer, 1.34 Kg gypsum (CaSO₄), and 0.89 Kg Micromax¹⁶. Initial pH¹⁷ was 5.7 and the final pH at the end of experiment was 5.9. Dormant rooted cuttings were planted on March 31, 1990 and placed under 30% shade for 10 days prior

¹³ Kocide 101, Griffin Corp., Valdosta, GA

¹⁴ Glidden Spread House - 100% Acrylic Latex No. 3600 White:
Glidden Coatings and Resins, Cleveland, OH

¹⁵ Zarn 300X, Zarn Inc., Reidsville, NC

¹⁶ Grace Sierra, Milpitas, CA

¹⁷ Corning Model 5 pH Meter, Medfield, MA

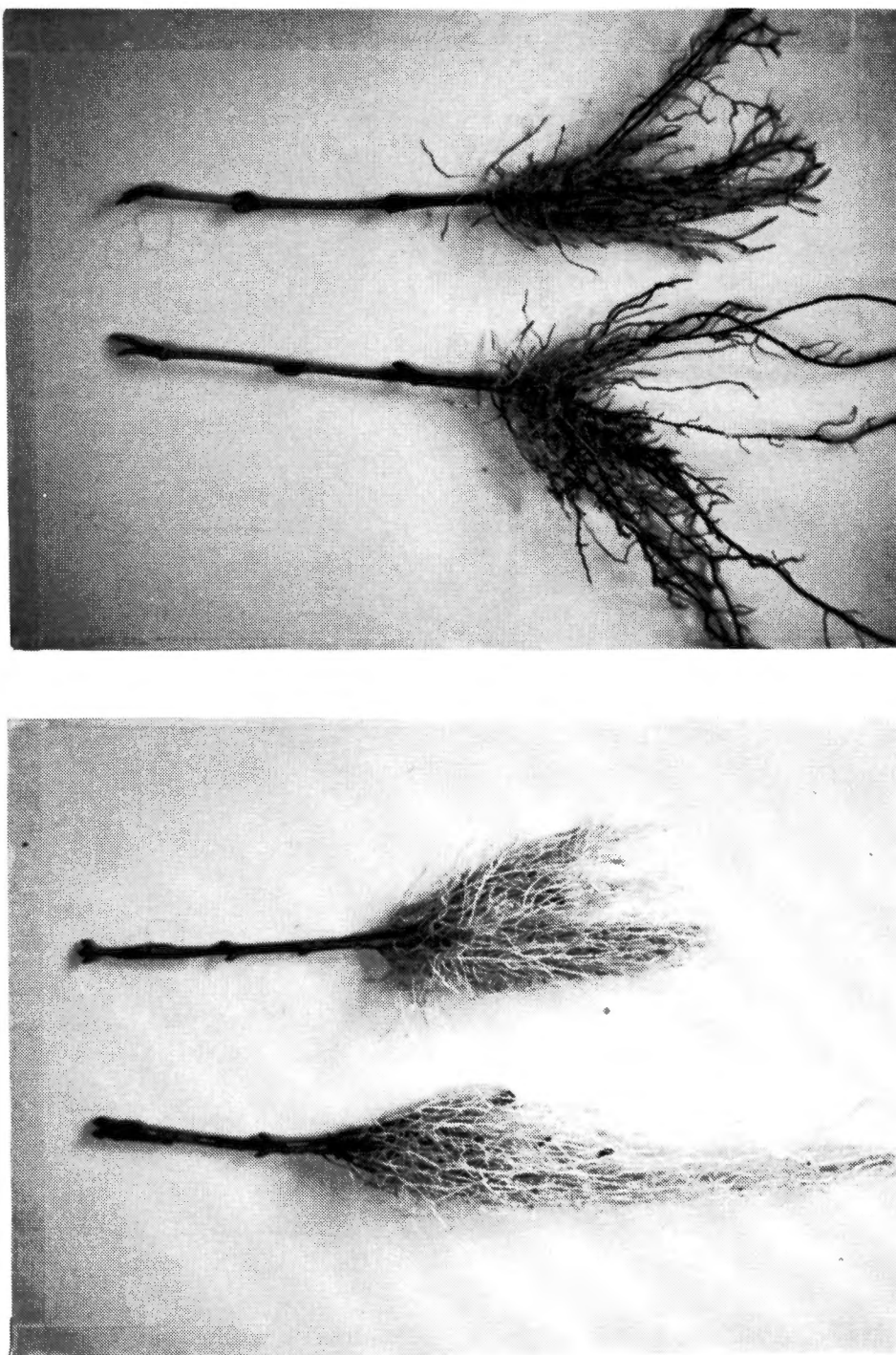


Figure 4. Bed-grown, rooted cuttings of euonymus (left photograph) and viburnum (right photograph). Untrimmed rooted cuttings on left of each photograph and roots trimmed to 8 cm on right of each photograph.

Table 4. Concentration and dilution schedule for incorporating cupric hydroxide (CH) in water prior to mixing with exterior acrylic latex paint (LP).

Treatment	Grams CH	Water (l)	LP (l)	Final Volume (l)	Grams CH/l LP (w:v)
1	0	0	0	0	0
2	0	0	2.0	2.0	0
3	100	0.090	2.0	2.12	47
4	200	0.140	2.0	2.20	90
5	500	0.350	2.0	2.52	185
6	1000	0.600	2.0	2.96	260

to moving them into full sun in the nursery research area. Plants were placed 45 cm on center.

Osmocote 18-6-12¹⁸ was topdressed at the rate of 12 grams per container 2 weeks after planting. Plants were liquid fertilized at 14 to 21 day intervals with Peter's 20-20-20¹⁹ at the rate of 200 ppm N (nitrogen). Watering was supplied by drip irrigation using Damm dribble rings.

On July 5, 1990, media temperature reached 43°C, which is considered to be a lethal root zone temperature by Whitcomb (81). All plants were moved into a 30% shade house. Heat stress on euonymus resulted in a 90% leaf drop, and those plants were eliminated from the experiment. Leaf drop on viburnum was about 10%, and plants were moved to a 30% shade house until termination of the experiment on September 5, 1990.

Subjective Root Analysis

On September 5, 1990, all viburnum plants were subjectively evaluated by 5 horticulturists for visible effects of chemical root pruning on the surface of the root ball. A rating scale of 1 to 5 was used, with 1 for no visible roots on the outside of the root ball; 2 for roots up to 2 cm in length; 3 for roots > 2 cm but < 5 cm in length; 4 for roots > 5 cm but < 10 cm in length, and 5 for roots > 10 cm in length.

¹⁸ Sierra Chemical Co., Milpitas, CA

¹⁹ W.R. Grace and Co., Fogelsville, PA

Counted Root Analysis

On September 6, 1990, 5 replications (30 plants total) were randomly selected and evaluated for effects of CH on reducing or eliminating the number of deflected roots which continued to grow after coming into contact with container walls. Roots were classified as either deflected or stubbed. Roots, growing on the outside of the container root ball, longer than 6.4 mm were classified as deflected and not controlled by CH. Roots stubbed at the surface of the root ball were classified as chemically root pruned.

Long Term Root Regeneration

On September 6, 1990, 5 replications, 30 plants plus those in the Counted Root Analysis (60 total) were stepped up to untreated, 12 l, black plastic containers²⁰. The same media and amendments were used as previously mentioned on page 27. Plants remained under 30% shade and were over-wintered in a poly-covered bow structure. Plants were removed from the over-wintering house April 20, 1991, and placed in the nursery research area 45 cm on center. Plants were liquid fertilized with Peter's 20-20-20²¹ at the rate of 200 ppm N every 4 to 5 days. On May 31, 1991 plants were removed from containers and subjectively evaluated by 5 horticulturists for root regeneration on the root ball surface using a rating scale of 1 for minimal root growth on the outside of the root ball, 2 for moderate and 3 for heavy.

²⁰ Zarn 1200X, Zarn Inc., Reidsville, NC

²¹ W.R. Grace and Co., Fogelsville, PA

Measured Root Regeneration

On September 18, 1990, 5 replications (30 plants) were placed into acrylic plexiglass growth trays to monitor root regeneration over a 32 day period. Growth trays were constructed of 6.44 mm acrylic plexiglass²². Inside dimensions of growth trays were matched to interior dimensions of the 3 l plastic container to allow for a snug fit of the plant within the tray. Aluminum foil covered the plexiglass and prevented light from reaching roots. At 2 day intervals, aluminum foil was removed and roots were measured (through the plexiglass) using a Digimatic caliper²³. Ten roots (300 total) were selected on each plant and monitored for 32 days. Initial root tip positions were marked on the plexiglass and growth was recorded as cumulative growth (in millimeters) every 2 days for 32 days.

Elemental Analysis

On October 20, 1990 root samples and the top third of the foliage (F) were removed from the 5 remaining replications for elemental analysis. Root tips (RT) in contact with container walls were removed in serial sections. RT designates the apical 5 mm root tip and S2 the next 5 mm. A third section, (S3), of the root system was composed of the first 2.5 cm below the soil surface. Roots which had been deflected and continued growing were not used in this sampling except for the untreated control where no CH was present.

²² Rohm and Haas, Philadelphia, PA

²³ Mitutoyo, Japan

The same root and leaf samples were also taken from the 5 replications in the plexiglass growth trays to compare elemental concentrations between roots and leaves of plants undergoing root regeneration to those in CH treated containers.

Root sections and leaves were placed in a Stabil-therm drying oven²⁴ at 105 °C for 48 hours. A mortar and pestle was used to grind both RT and S2. In order to grind F and S3 it was necessary to use a Wiley Mill²⁵ with a #20 mesh screen. Due to weight requirements (0.5 grams per sample) for elemental analysis it was necessary to pool both RT and S2 samples into their respective treatments. Since those samples were pooled, so were S3 and F samples. Elemental analysis was performed by MMI, Inc.²⁶.

Histology Studies

Root tips, 3 mm length, were removed from plants used in the elemental analysis studies, were fixed in CrAF III (60) and 50% FAA (60), degassed in a vacuum chamber, dehydrated in a graded 2-propanol series and embedded in Polyfin^{™27}. Sections, 10µm, were cut on a rotary microtome, expanded on a water pool on a glass slide and dried on a warming tray. Sections were stained in 0.01% toluidine blue (borax buffer, pH 9.0) for

²⁴ Blue M Electric, Blue Island, IL

²⁵ Thomas Co., Philadelphia, PA

²⁶ Athens, GA

²⁷ Polysciences Inc., Warrington, PA

30 minutes (30). Stained sections were deparaffinized in Micro-Clear™²⁸ and cover glasses were affixed with EUKITT®²⁹ mounting reagent.

Results and Discussion

Data were analyzed with a GLM procedure using Statistical Analysis Software³⁰. Means were separated at the 5% level of significance.

Subjective Root Rating

Analysis of subjective evaluations showed all levels of CH had fewer roots on the root ball surface than control and LP only treatments. As CH levels increased, surface roots decreased (Table 5). Roots which developed on the west side of the root ball were destroyed due to high temperatures from solar heating. Therefore all ratings refer to 'best side' (heaviest rooted side) of the root ball.

Counted Root Analysis

CH reduced the number of deflected roots continuing to grow after contact with container walls by 55% to 92% (Table 6).

CH at 90 g/l chemically root pruned more roots than any other CH rate, control or LP only treatment (Table 6). Roots in the control treatment cannot be considered as

²⁸ Micron Diagnostics Inc., Baltimore, MD

²⁹ Calibrated Instruments, Inc., Ardsley, NY

³⁰ SAS 6.03, SAS Institute, Inc., Cary, NC

Table 5. Mean rating of root growth on root ball surface of *Viburnum tomentosum plicatum* 'Mariesii' grown 5 months in 3 l containers painted with cupric hydroxide (CH)/exterior acrylic latex paint (LP) on interior walls.

Treatment	Grams CH/LP	Mean Rating ^z
1 Control	0	3.8 a
2 LP Only	0	3.8 a
3 CH + LP	47	2.9 b
4 CH + LP	90	2.4 c
5 CH + LP	185	2.0 d
6 CH + LP	260	1.5 e

GLM Procedure, Duncan's Multiple Range Test.

^z Means within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

Table 6. Mean number of deflected and chemically root pruned roots of Viburnum tomentosum plicatum 'Mariesii' grown 5 months in containers painted with cupric hydroxide (CH)/exterior acrylic latex paint (LP) on the inside walls.

Treatment	Grams CH/LP	# of deflected roots ^z	# of roots chemically pruned ^z
1 Control	0	112.4 a	1.4 c
2 LP Only	0	87.4 ab	11.8 c
3 CH + LP	47	61.6 b	49.4 b
4 CH + LP	90	14.8 c	96.8 a
5 CH + LP	185	12.4 c	71.0 ab
6 CH + LP	260	9.2 c	61.6 b

ANOVA Procedure, Waller-Duncan K-ratio T test.

^z Means within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

stubbed since they would eventually be deflected by the wall as they elongate. At the 260 g/l CH treatment, stubbed root tips were thicker, club-like and necrotic at point of contact with the container wall. There seemed to be a stimulatory effect on development of lateral branching roots behind root tips at the highest CH level, however these roots appeared to be short lived. After contacting treated walls they took on the same characteristics as the first root tip.

Long Term Root Regeneration

Table 7 shows mean subjective ratings of viburnum rooted cuttings 8 months after transplanting from CH treated containers. Only treatment 3 (47 g/l CH) was different from treatment 6, 260 g/l CH based on a rating scale of 1 for minimal roots on the outside of the root ball, 2 for moderate and 3 for heavy. Root development occurred over the entire container root ball surface during early spring while plants were still in the over-wintering house. After plants were placed in full sun, root growth stopped on the sunny west side of the container. Therefore the rating scale was applied to 'the best side' of the root ball.

Measured Root Regeneration

After 32 days, plants grown in the 90 g/l CH treatment reached the same level of root growth as control plants. Based on individual treatment data a statistical growth curve was derived to model root regeneration of each CH treatment (Figure 5) utilizing

Table 7. Mean rating of root growth on root ball surface of Viburnum tomentosum plicatum 'Mariesii' grown for 5 months in 3 l cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers, and 8 months in untreated 12 l containers.

Treatment	Grams CH/LP	Rating ^z
1 Control	0	1.84 ab
2 LP Only	0	1.86 ab
3 CH + LP	47	1.93 a
4 CH + LP	90	1.74 ab
5 CH + LP	185	1.76 ab
6 CH + LP	260	1.58 b

GLM Procedure, Duncan's Multiple Range Test.

^z Means followed by the same letter are not significantly different at $\alpha = 0.05$.

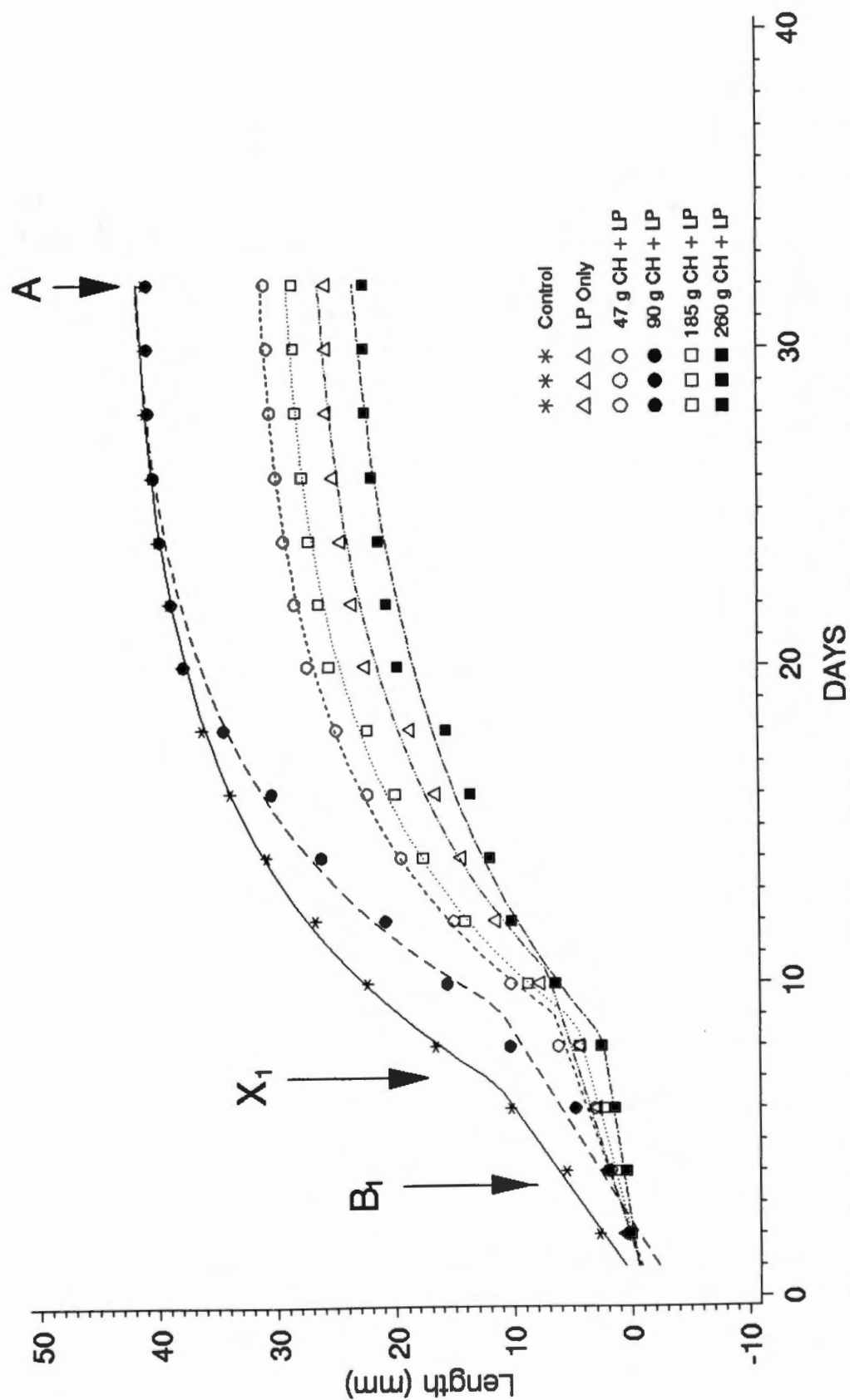


Figure 5. Root regeneration growth curve of viburnum, chemically root pruned with CH/LP treatments, after 32 days in untreated plexiglass growth trays. B_1 = slope, X_1 = days to beginning of exponential growth and A = asymptote.

3 parameters: 1) slope (B_1) representing linear growth rate, 2) change from linear growth to an exponential growth pattern (X_1) and 3) an asymptotic curve (A). Figure 5 is a combination of linear versus non-linear growth, where a growth curve is fit through the means and analyses are through the means of the curve.

Under normal growing conditions many plant organs exhibit an S - shaped, sigmoid, growth curve which is considered to be typical (59). Three phases characterize a sigmoid growth curve: 1) a logarithmic phase, 2) a linear phase and 3) a senescence phase.

Initial growth of roots in all treatments was linear, and the equation $Y = a + bX$ was used to calculate the slope (B_1). Roots from control plants, already in a linear growth pattern, grew faster initially, and the slope was significantly greater than any other treatment (Table 8). CH at 260 g/l significantly reduced initial root growth compared with all other treatments.

Treatment effects were evident when root growth changed from a linear growth pattern to an exponential (X_1) growth pattern (Table 8). Control and 97 g/l CH began exponential growth at the same time. LP had an inhibitory effect on regeneration of roots that were in contact with container walls treated with paint.

An asymptote (A), is a line approached by a curve, where Y approaches zero and X increases without limit (42). In this case Y is the length of the root when root growth has leveled off and X is number of days. No significant differences occurred between the 90 g/l CH treatment and control plants (Table 8). All other treatments had significantly lower root lengths. Mean regenerated root growth was reduced by approximately 25%

Table 8. Statistical root growth curve parameters established for slope (B_1), exponential growth (X_1) and asymptote (A) for Viburnum tomentosum plicatum 'Mariesii' grown in cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers for 5 months and untreated plexiglass growth trays for 32 days.

Treatment	Grams CH/LP	Slope (B_1) ^z	Days to (X_1) ^z	Length (mm) of roots at Asymptote (A) ^z
1 Control	0	1.52 a	7.20 d	42.8 a
2 LP Only	0	0.57 c	10.12 a	28.2 d
3 CH + LP	47	0.64 c	8.22 bcd	33.0 b
4 CH + LP	90	0.86 b	7.66 cd	43.8 a
5 CH + LP	185	0.55 c	8.47 bc	30.2 c
6 CH + LP	260	0.27 d	9.02 b	25.8 e

GLM Procedure, Duncan's Multiple Range Test.

^z Means within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

in the 47 g/l CH treatment, 31%, 36% and 41% in the 185 g/l, LP only and 260 g/l CH treatments, when compared to control and 90 g/l CH treatments

After transplanting from treated containers into plexiglass growth trays, 100% root regeneration occurred within 2 days in control plants: 88% in 4 days for the LP only treatment; 92% in 4 days, 98% in 4 days, 76% in 4 days and 92% in 6 days for the 47, 90, 185, and 260 g/l CH treatments respectively.

Elemental Analysis

Phosphorous (P), potassium (K), calcium (Ca) and magnesium (Mg) levels (Table 9) were within acceptable ranges for dry matter in all plant tissue samples (7, 8, 25, 35, 47). CH effectively inhibited root growth on viburnum. However, there appeared to be no decrease in uptake of P or K as a result of root inhibition as suggested by Foy *et al.* (20).

Manganese (Mn) levels were higher in RT, S2 and S3, when in contact with LP only and CH treated walls (Table 9). Conversely, after 32 days of growth in untreated containers, Mn levels dropped in all root tissues. Mn levels were consistent in roots of the control treatment as well as in foliage of all plants with roots in contact with CH treated container walls and those which were grown for 32 days in untreated containers (Table 9). Chemically, Mn shows properties similar to Mg, Ca and the heavy metals Zn and Fe. According to Mengel *et al.* (47), Mn may antagonistically reduce uptake of Fe.

RT and S3 sections of viburnum roots growing in CH treated containers had lower concentrations of Fe compared with those grown in untreated containers (Table 9). Fe

Table 9. Concentration analyses (ppm) of selected elements in root tissues of *Viburnum tomentosum plicatum* 'Mariesii' grown in cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers for 5 months (+) and after 32 days in untreated containers (-).

Element		CH/LP Treatments				
Code ^z	Control	LP Only	47 g/l	90 g/l	185 g/l	260 g/l
P						
RT +	4700	4700	4200	5300	4400	5300
RT -	5400	4400	4800	5500	5800	4700
S2 +	5100	6000	6900	6900	7400	5500
S2 -	10,400	9900	6700	8900	10,500	8700
S3 +	2400	2000	1800	1700	2300	2500
S3 -	4200	3400	4000	3600	4000	2800
F +	4300	3200	2700	3400	3100	3200
F -	3100	3900	4300	4500	4700	4400
K						
RT +	12,900	10,900	10,100	13,100	10,200	9900
RT -	11,700	6900	10,600	11,800	12,300	9600
S2 +	16,700	16,800	22,000	19,500	23,900	16,600
S2 -	33,200	35,400	16,100	35,100	36,500	32,000
S3 +	3200	2200	2200	2300	3400	3100
S3 -	4000	3900	4300	3700	4200	3800
F +	20,400	16,100	16,600	19,700	17,500	18,300
F -	16,600	24,300	24,400	24,000	24,700	22,700

Table 9. (continued)

Element		CH/LP Treatments				
Code ^z	Control	LP Only	47 g/l	90 g/l	185 g/l	260 g/l
Ca						
RT +	7700	7400	6600	6700	7400	7300
RT -	8000	8100	7500	7800	9700	8700
S2 +	6300	6000	6100	6300	5300	6800
S2 -	4800	5000	7500	4000	4700	4600
S3 +	8100	8000	7600	5700	9400	8100
S3 -	11,600	9600	11,700	10,300	12,900	10,500
F +	9700	12,700	11,200	11,600	12,800	11,400
F -	12,600	11,100	11,400	10,200	11,100	11,200
Mg						
RT +	2800	2300	2300	3000	2400	2500
RT -	2800	1900	2100	2900	2400	2400
S2 +	3400	3300	4300	3900	4800	3500
S2 -	6000	5600	3300	5300	6100	5100
S3 +	1300	1200	1100	1100	1400	1500
S3 -	1900	1500	1600	1700	1700	1700
F +	1600	2000	1800	1600	1900	1900
F -	2000	1800	1700	1600	1700	1700

Table 9. (continued)

Element		CH/LP Treatments				
Code ^z	Control	LP Only	47 g/l	90 g/l	185 g/l	260 g/l
Mn						
RT +	126	318	216	307	193	181
RT -	123	128	111	192	112	201
S2 +	104	410	236	258	310	242
S2 -	100	103	244	124	144	159
S3 +	185	542	339	324	254	268
S3 -	168	194	160	257	140	168
F +	172	175	182	214	202	230
F -	165	160	169	196	183	220
Fe						
RT +	119	129	120	119	109	77
RT -	123	135	146	163	159	109
S2 +	114	138	165	136	158	131
S2 -	105	102	118	132	123	110
S3 +	163	103	102	149	152	170
S3 -	189	195	210	198	247	219
F +	186	208	232	220	250	228
F -	200	261	247	213	279	245

Table 9. (continued)

Element		CH/LP Treatments				
Code ^z	Control	LP Only	47 g/l	90 g/l	185 g/l	260 g/l
Cu						
RT +	14	16	103	109	121	312
RT -	26	12	25	48	36	46
S2 +	17	22	77	75	100	192
S2 -	60	24	28	73	132	152
S3 +	7	5	5	4	8	8
S3 -	25	9	13	12	12	9
F +	6	2	2	4	5	8
F -	2	3	6	5	4	7
Zn						
RT +	92	87	112	174	188	177
RT -	71	74	109	130	150	118
S2 +	65	78	175	174	265	293
S2 -	80	69	136	169	205	258
S3 +	47	46	31	31	41	48
S3 -	92	63	85	72	66	52
F +	81	98	75	90	85	90
F -	84	81	91	95	77	93

^z RT = Root tip (5mm); S2 = First 5 mm behind root tip;
 S3 = First 2.5 cm of main root below the soil surface;
 F = Foliage.

levels in S2 sections from treated containers were higher than those in untreated containers. A gradient existed between RT and F across all treatments and tissue sections (Table 9). Concentrations of Fe approximately doubled between RT and F. Whether depression of Fe in the root tissue sections of plants with roots in contact with CH treated walls is a result of inhibition by Mn or Cu cannot be determined from this data.

Roots in contact with CH treated walls had Cu levels higher than roots regenerated in untreated containers for 32 days (Table 9). Based on Mengel and Kirby (47), the normal range of Cu in dry plant matter is from 2 - 20 ppm. Bonner and Varner (7), stated that 6 ppm Cu is an adequate level in plant material. High levels of Cu decreased rapidly, from 312 ppm to 46 ppm after 32 days in untreated containers, in RT sections from the 260 g/l CH treatment. Even though Cu levels rapidly decreased, no root regeneration was observed from root tips. Regeneration occurred in the form of lateral and secondary branching roots immediately behind RT. Symptoms of mild Cu toxicity (36) were evident at the 185 g/l CH treatment. Roots were beginning to thicken and became darker in color. At 260 g/l CH roots in contact with treated container walls were thick, tips were club-like and root tissue was becoming brittle and necrotic. Cu levels in S2 root sections grown for 32 days in untreated containers decreased less than RT and S3 in the same plant (Table 9). Foliage levels of Cu were acceptable in all treatments (Table 9).

Zn levels were within the normal range of 100 ppm in dry matter (47) of all RT, S2, S3 and F tissues of control and LP only treatments, as well as S3 and F tissues of both treated and untreated containers (Table 9). Concentration of Zn in RT and S2

tissues in both treated and untreated containers were higher than the normal range. Zn uptake did not appear to be inhibited by Cu in this study, contrary to results by Schmid *et al.* (62) with barley roots, and by Bowen (9) with sugar cane leaves.

Histology Studies

Over 400 slides of root tips from all treatments were prepared and evaluated in this study. All specimens were prepared as longitudinal, serial sections. Those specimens prepared in 50% FAA were hard, and tore when sectioned on the rotary microtome. These results are based on specimens prepared in CrAF III.

Root tips from untreated containers (Figure 6) showed normal growth and development with cellular structures clearly defined. In the 90g/l CH treatment (Figure 6) the cortex and vascular cylinder were more mature, and ergastic substances were more prevalent in the cortex. In the 260 g/l CH treatment (Figure 6) roots became thicker and club-like, with root tip structure appearing callus-like with no visible cellular definition within the root tip or root cap area. As little as 90 g/l CH was sufficient for chemical root pruning of viburnum. No visual signs of Cu toxicity were observed at levels up to 90 g/l. At higher levels, root tips were visibly darker and thicker.

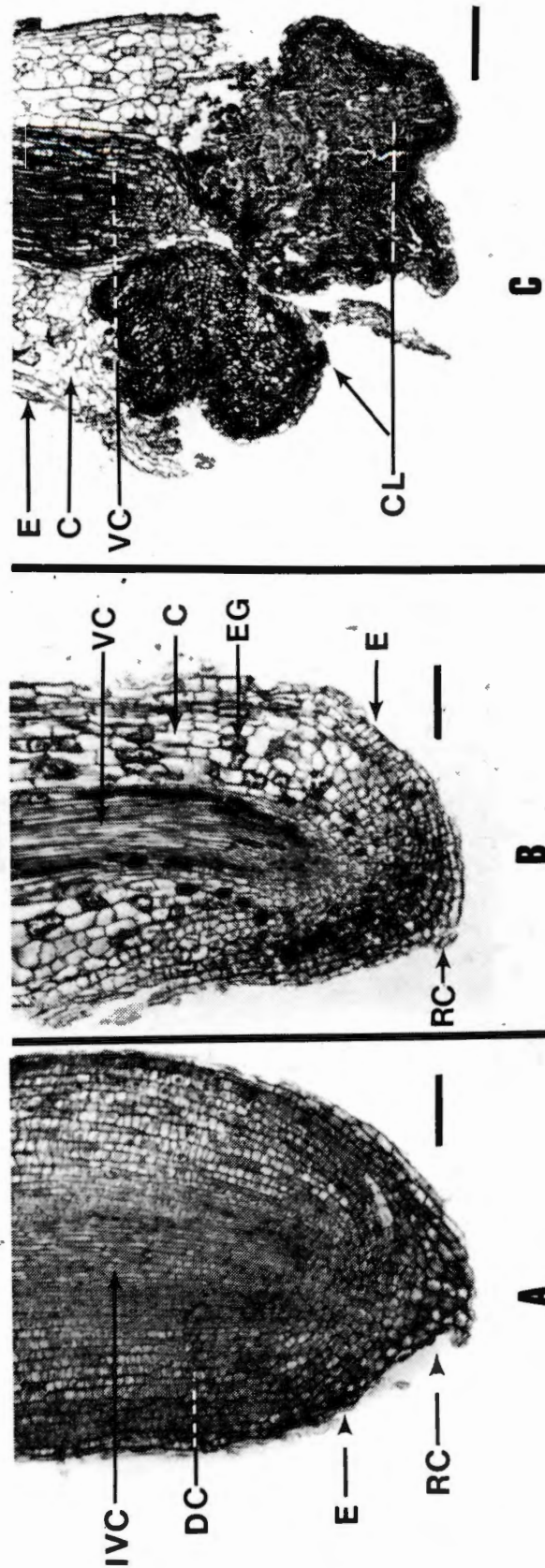


Figure 6. Microphotographs of medial, longitudinal root tip sections of viburnum. Plants were grown for 5 months in 3 l. plastic containers. (A) Normal root tip grown in untreated container. RC, root cap; E, epidermis; DC, developing cortex; IVC, immature vascular cylinder. Bar = 100 μ m. (B) Root tip in contact with container wall painted with 90 g/l cupric hydroxide in latex paint. RC, root cap; E, epidermis; EG, ergastic substances; C, cortex; VC, maturing vascular cylinder. Bar = 100 μ m. (C) Root tip in contact with container wall painted with 260 g/l cupric hydroxide in latex paint. CL, callus-like mass replacing root cap; E, epidermis; C, cortex; VC, mature vascular cylinder. Bar = 300 μ m.

CHAPTER V

EFFECTS OF $\text{Cu}(\text{OH})_2$ ON SECONDARY BRANCHING OF ROOTS

Introduction

Saul (61) reported that roots could be safely root pruned and possibly remain in smaller containers, without detrimental effects, by using copper paint. Copper concentration was shown to be important for chemical root pruning effects over an extended period of time (57, 58). Lower concentrations (25 g/l) proved inadequate.

Some reports showed vigorous growth of transplants removed from copper treated containers (11, 13, 45), and others stated that copper treatments did not affect height, caliper or FW and DW of roots or shoots (58). Within the root systems of copper treated plants Nussbaum (50) reported an increase in secondary and tertiary roots.

An experiment was performed to determine the effects of different rates of CH/LP treatments on secondary branching of euonymus plants grown in 0.9 l pots for an extended period of time.

Materials and Methods

This experiment was arranged as a randomized complete block design with 6 treatments and 5 replications with 5 root samples per treatment/replication.

Procedures for preparation and application of the CH mixture and type of media were detailed in CHAPTER IV, page 26.

On May 1, 1990, dormant rooted cuttings of euonymus were removed from cold storage, 4 °C, and 30 uniform plants selected. Roots were pruned to 8 cm and shoots to 6 cm. Rooted cuttings were planted in treated, 10 cm, 0.9 l, black plastic containers³¹ and placed on raised benches, 20 cm on center, in a double-layer poly-covered greenhouse with 30% shade. Shade cloth was removed October 15, 1990. Average day/night temperatures were 23.3 °C/15.6 °C $\pm$ 1.7 °C. Plants remained in the greenhouse for the entire experiment. Plants were liquid fertilized with Peter's 20-20-20³² at the rate of 200 ppm at 5 to 7 day intervals. All watering was done by hand.

Plant heights were recorded on November 1, 1990, 6 months after the experiment was initiated. Five roots in contact with treated container walls were randomly selected from each plant. The distance (mm) between the root tip and each lateral branching root was recorded for the first 2.5 cm of the main root. There were no more than 5 lateral branch roots for any root sample. Measurements were made using a Digimatic caliper³³.

One plant from each treatment was randomly selected and media gently washed from the root ball. Plants were then suspended in a tank of water, allowing roots to float freely. Root systems were visually evaluated for formation and development.

³¹ Classic 100, Nursery Supplies Inc., Fairless Hills, PA

³² W.R. Grace and Co., Fogelsville, PA

³³ Mitutoyo, Japan

Media was washed from root systems of the remaining 24 plants (30 total). Shoots were removed from roots at soil level and FW taken on both. Weights were recorded on a Model 1200P balance³⁴. Plants were then placed in a Stabil-therm drying oven³⁵ for 96 hours at 105 °C, removed and DW recorded.

Data were analyzed using Statistical Analysis Software³⁶. Means were separated at the 5% level of significance.

Results and Discussion

No significant treatment effects were observed for the height of euonymus.

Table 10 shows that the mean number of lateral roots occurring within the distal 2.5 cm of root tips increased as the concentration of CH increased. This was also true for the distance to the first branch root. Mean distance between roots decreased in the LP only treatment, and as CH concentrations increased.

Table 11 shows plants grown in containers treated with 90 g/l CH had shoot fresh weights 10% greater than control plants; 25% greater than 260 g/l CH and 31% greater than the LP only treatment. No significant differences occurred between control, 47 g/l CH and 185 g/l CH treatments in FW/DW of shoots. Treatment 4, 90 g/l CH, appeared to have a stimulatory effect on shoot growth, while LP and 260 g/l CH treatments appeared inhibitory.

³⁴ American Scientific Products, McGraw Hill, IL

³⁵ Blue M Electric, Blue Island, IL

³⁶ SAS 6.03, SAS Institute, Inc., Cary, NC

Table 10. Comparison of mean number of lateral branching roots within the first 2.5 cm of the root tip, and mean mm distance between first 5 branch roots of Euonymus alatus 'Compacta' grown 6 months in cupric hydroxide(CH)/exterior acrylic latex paint (LP) treated containers. No more than 5 branch roots occurred at any treatment level.

Treatment	Grams CH/LP	# of lateral roots ^z	mm to first branch root ^z	mm between branch roots ^z
1 Control	0	2.3 c	20.5 a	10.5 a
2 LP Only	0	4.0 b	13.5 b	5.9 b
3 CH + LP	47	4.8 a	4.8 c	3.9 c
4 CH + LP	90	5 a	3.8 c	3.7 c
5 CH + LP	185	5 a	3.1 c	3.4 c
6 CH + LP	260	5 a	2.0 d	1.8 d

GLM Procedure, Duncan's Multiple Range Test.

^z Means within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

Table 11. Grams fresh weight (FW) and dry weight (DW) of *Euonymus alatus* 'Compacta' shoots and roots of plants produced in cupric hydroxide (CH)/exterior acrylic latex paint (LP) treated containers for 6 months.

Treatment	g FW shoots ^z	g DW shoots ^z	g FW roots ^z	g DW roots ^z
1 Control	39.9 b	16.2 b	76.1 b	23.2 b
2 LP Only	30.4 c	13.4 c	45.4 d	13.2 d
3 CH + LP	38.7 b	16.2 b	66.3 c	19.3 c
4 CH + LP	43.8 a	18.6 a	90.1 a	27.7 a
5 CH + LP	38.9 b	17.1 ab	51.0 d	15.7 d
6 CH + LP	32.9 c	13.5 c	43.2 d	12.4 d

GLM Procedure, Duncan's Multiple Range Test.

^z Means within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

Table 11 shows mean FW of root systems from the 90 g/l CH treatment was significantly greater (16%) than control plants, and 49% greater than the LP only, 185 g/l CH and 260 g/l CH treatments. DW were exactly correlated to FW (Table 11).

Visual observations of root systems submerged in a glass water tank showed control plants had a good fibrous root system within the root ball (Figure 7). However, the long roots floating freely around the container ball are the same roots coiled around the inside walls of the pot. Treatments 4 and 6, 90 g/l CH and 260 g/l CH, appeared to be similar in characteristics. In Table 10, mean distance between roots in the first 2.5 cm of the root tip of the 260 g/l CH treatment was 1.8 mm. Closer examination of roots in treatment 6, showed there was a gap of 2 to 3 cm between branching roots behind the root tip and where branching started again (Figure 8). This same characteristic was observed in the 185 g/l CH treatment, but not in other CH treatments where the secondary branching was continuous along the root behind root tips. Signs of Cu toxicity such as thickening of roots, swelling of root tips, or darkening of roots were only observed in treatment 6, 260 g CH/LP. Absence of secondary branching may account for differences in FW and DW between the treatments.

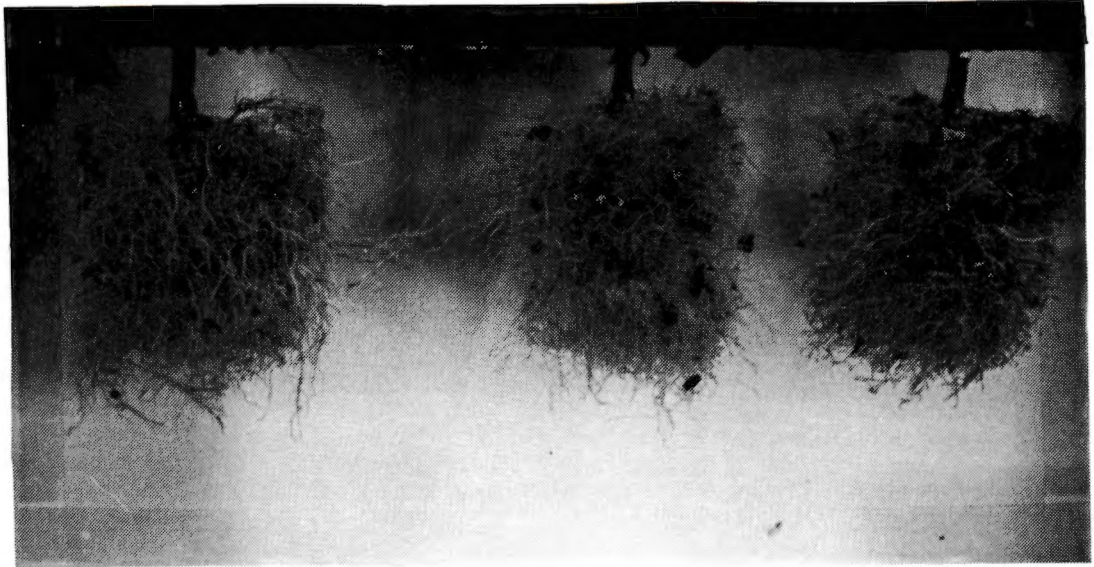


Figure 7. Cleaned euonymus root balls, suspended in water, grown 6 months in CH/LP treated containers. Control, 0 CH/O LP, is on left, 90 g CH/LP in center and 260 g CH/LP on right.

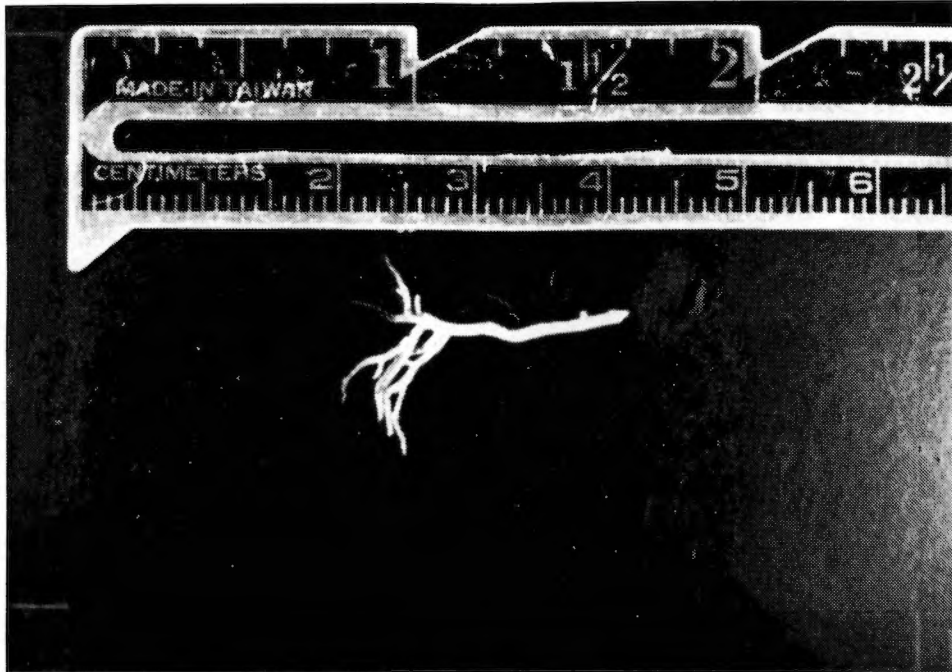


Figure 8. Multiple branching behind root tips of euonymus in contact with 260 g CH/LP after 6 months in treated containers. Note root tips which were in contact with treated container walls, thickening of roots and lack of branching behind first group of secondary roots.

CHAPTER VI

CONCLUSIONS AND SUMMARY

General Conclusions

EFFECTS OF $\text{Cu}_4\text{H}_6\text{O}_{10}\text{S}$ AS A CHEMICAL

ROOT PRUNING AGENT

TCS from 47 to 228 g/l LP was effective in reducing the number of deflected roots on sawtooth oak seedling when roots came into contact with tube walls coated with this material. There were fewer fibrous roots on plug surfaces in all TCS treatment levels compared with both controls.

Three weeks after transplanting, plant height and regeneration of lateral roots from tap roots were increased by the 47 g/l TCS treatment.

EFFECTS OF $\text{Cu}(\text{OH})_2$ AS A CHEMICAL

ROOT PRUNING AGENT

CH in combination with LP was effective in chemically root pruning rooted viburnum cuttings. Treatments containing 90, 185 and 260 g/l CH reduced the number of deflected roots continuing to grow after contacting CH treated container wall surfaces.

Regeneration of root growth after removal from treated containers was slower compared to control plants, but after 32 days, roots from 90 g/l CH treatment were equal

to control treatment roots. Roots from 185 and 260 g/l CH treatments were 35% shorter after the same period of time.

Cu concentration was highest in root tips contacting the CH treated walls and decreased rapidly from root tip to foliage tissue. No visual signs of Cu toxicity appeared on foliage. Foliage Cu levels were normal in all treatments. CH treatments did not appear to adversely affect other mineral elements within the plant.

Roots in contact with container walls treated with CH were thicker and darker in color. At the highest rate, root tips became club-like and necrotic. Histological evaluation revealed a callus-like appearance of the root tip and apical meristem area, with no apparent cellular organization. Root regeneration was by secondary branching behind the root tip at the highest CH treatment.

EFFECTS OF $\text{Cu}(\text{OH})_2$ ON SECONDARY BRANCHING OF ROOTS

Secondary branching of roots was increased when roots came into contact with the treated container walls in all CH mixtures and LP only treatments. FW/DW of roots in the 90 g/l CH treatment were 53% greater than the worst treatment, 260 g/l CH and 16% greater than the untreated control. While LP was effective in promoting secondary branching, it was inhibitory in FW and DW of shoots and roots.

Summary

These experiments were designed to test the effects of alternative copper compounds, TCS and CH in LP, as chemical root pruning agents on container grown woody ornamental plants. Concentration rates were from 47 to 260 grams copper per liter of paint.

Both TCS and CH in combination with LP were effective as chemical root pruning agents, aiding in elimination of root coiling and kinking in the container. As little as 90 g/l TCS or CH was sufficient for chemical root pruning in these experiments. No visual signs of copper toxicity were observed at levels up to 90 g/l. Foliage showed no signs of chlorosis, and roots showed no signs of thickening or becoming darker.

Increased secondary root growth was observed in all plants tested. Plants with larger, more fibrous root systems provide a larger root surface area for the absorption of water and minerals, and should respond well when set into the landscape.

LIST OF REFERENCES

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1. Addoms, R. M. 1946. Entrance of water into suberized roots of trees. *Plant Physiol.* 21:109-111.
2. Appleton, Bonnie Lee and Carl E. Whitcomb. 1983. Effects of container size and transplanting date on growth of tree seedlings. *J. Environ. Hort.* 1:89-93.
3. Arnold, M. A. 1987. Cupric carbonate modification of Quercus rubra and Fraxinus pennsylvanica root systems and implications for production transplant. M.S. Thesis. The Ohio State University.
4. Arnold, M. A. and Daniel K. Struve. 1989. Cupric carbonate controls green ash root morphology and root growth. *Hortscience* 24(2):262-264.
5. Arnold, M. A. and Daniel K. Struve. 1989. Growing green ash and red oak in CuCO₃ treated containers increases root regeneration and shoot growth following transplant. *J. Amer. Soc. Hort. Sci.* 114(3):402-406.
6. Barnett, J. P. and J. M McGiveray. 1974. Copper screen controls root growth and increases survival of containerized southern pine seedlings. *Tree Planters Notes.* 25(2):11-12.
7. Bonner, J. and J. E. Varner. 1965. *Plant biochemistry.* Academic Press, New York.
8. Bould, C. 1970. The nutrition of tree crops. In: L. C. Luckwill and C. V. Cutting (eds.) *Physiology of tree crops.* Academic Press. New York, New York.
9. Bowen, J. E. 1969. Absorption of copper, zinc and magnesium by sugar cane tissue. *Plant Physiol.* 44:255-261.
10. Brady, Nyle C. 1974. *The nature and properties of soil.* 8th Edition. McMillan Press, New York.
11. Burdett, A.N. 1978. Control of root morphogenesis for improved mechanical stability of container-grown lodgepole pine. *Can. J. For. Res.* 8(4):483-486.
12. Burdett, A. N. 1981. Box-pruning the roots of container-grown tree seedlings. *Proceedings of the Canadian Containerized Tree Seedling Symposium.* p.203-206.

13. Burdett, A. N. and P. A. F. Martin. 1982. Chemical root pruning of coniferous seedlings. *HortScience* 17(4):622-624.
14. Cataldo, D. A. and R. E. Wildung. 1978. Soil and plant factors influencing the accumulation of heavy metals by plants. *Environ. Health Perspectives* 27:149-159.
15. Chong, C., G. P. Lumis, R. A. Cline and H. J. Reismann. 1987. Growth and chemical composition Populus deltoides x nigra grown in field-grow fabric containers. *J. Environ. Hort.* 5:45-48.
16. Daniels, R. R., B. E. Stuckmeyer and L. A. Peterson. 1972. Copper toxicity in Phaseolus vulgaris L. as influenced by iron nutrition: an anatomical study. *J. Amer. Soc. Hort. Sci.* 9:249-254.
17. Davidson, H. and R. Mecklenburg. 1981. Nursery management administration and culture. Prentice-Hall, Englewood Cliffs, New Jersey.
18. DeWald, L. E. and P. P. Feret. 1987. Changes in loblolly pine root growth potential from September to April. *Can. J. For. Res.* 17:635-643.
19. Doug, H. and A. N. Burdett. 1986. Chemical root pruning of Chinese pine seedlings raised in cupric sulfate impregnated paper containers. *New Forests* 1(1):67-73.
20. Foy, C. D., R. L. Chaney and M. C. White. 1978. The physiology of metal toxicity in plants. *Annu. Rev. Plant Physiol.* 29:511-566.
21. Fuller, D. L. and W. A. Meadows. 1987. Root and top growth response of five woody ornamentals species to field-grow containers, bed height and trickle irrigation. *Proc. SNA Res. Conf.* 32:148-151.
22. Furuta, T., W. C. Jones, W. Humphrey and T. Mock. 1972. Chemically controlling root growth in containers. *Calif. Agr.* 26(2)10-11.
23. Geisler, D. and D. C. Ferree. 1984. Response of plants to root pruning. *Hort. Rev.* 6:155-188.
24. Geisler, Dagmar and David C. Ferree. 1984. The influence of root pruning on water relations, net photosynthesis, and growth of young 'golden delicious' apple trees. *J. Amer. Soc. Hort.* 109:827-831.
25. Gilliam, C. H. and E. M. Smith. 1980. Fertilization of container-grown nursery stock. Cooperative Extension Service. The Ohio State University. Bul. 658. p.18.

26. Gilman, Edward F. 1988. Tree root spread in relation to branch dripline and harvestable root ball. *HortScience* 23:351-353.
27. Harris, Richard W. 1983. *Arboriculture - care of trees, shrubs and vines in the landscape*. Prentice-Hall, Inc., New Jersey.
28. Harris, Richard W., W. B. Davis, N. W. Stone and Dwight Long. 1971. Root pruning improves nursery tree quality. *J. Amer. Soc. Hort. Sci.* 96:105-108.
29. Hill, J. M. 1973. The changes with age in the distribution of copper and some copper containing oxidase in red clover (*Trifolium pratense* L. cv Dorset Marlgrass). *J. Exp. Bot.* 24:525-536.
30. Humason, G. L. 1972. *Animal tissue techniques*. W. H. Freeman Co., San Fransico, California.
31. Ingram, Dewayne L., Uday Yadav and Cathy Neal. 1986. Research with field-grow containers in Florida. *Proc. SNA Res. Conf.* 31:94-97.
32. Kelly, R. J. and B. C. Moser. 1983. Root regeneration of *Liriodendron tulipifera* in response to auxin, stem pruning, and environmental conditions. *J. Amer. Soc. Hort. Sci.* 108:1085-1090.
33. Kozlowski, T. T. 1971. *Growth and development of trees*, vol II. Academic Press, Inc. New York, New York.
34. Kramer, P. J. and H. C. Bullock. 1966. Seasonal variation in the proportions of suberized and unsuberized roots of trees in relation to absorption of water. *Amer. J. Bot.* 53:200-204.
35. Kramer, P. J. and T. T. Kozlowski. 1979. *Physiology of woody plants*. Academic Press, New York, New York.
36. Kuhns, L. J. and T. D. Sydnor. 1976. Copper toxicity in woody ornamentals. *J. Arbor.* 2(4):68-72.
37. Larson, M. M. 1970. Root regeneration and early growth of red oak seedlings: influence of soil temperature. *For. Sci.* 16(4):442-446.
38. Larson, M. M. 1984. Seasonal planting, root regeneration, and water deficits of Austrian pine and arborvitae. *J. Environ.Hort.* 2:33-38.

39. Larson, M. M. 1980. Effects of atmospheric humidity on zonal water stress on initial growth of planted northern red oak seedlings. *Can. J. For. Res.* 10:549-554.
40. Lathrop, J. K. and R. A. Mecklenburg. 1971. Root regeneration and root dormancy in taxus spp.. *J. Amer. Soc. Hort. Sci.* 96:111-114.
41. Lee, C. T. and W. P. Hackett. 1976. Root regeneration of transplanted Pistacia chinensis Bunge seedlings at different growth stages. *J. Amer. Soc. Hort. Sci.* 101:236-240.
42. Little, Thomas M. and F. Jackson Hills. 1977. *Agricultural experimentation: design and analysis.* John Wiley and Sons, New York, New York.
43. Lyr, H. and G. Hoffman. 1967. Growth rate and periodicity of tree roots. *Int. Rev. For. Res.* 2:181-235.
44. McDonald, S. E., R. W. Tinus and C. P. P. Reid. 1981. Root development control measures in containers: recent findings. *Proceedings of the Canadian Containerized Tree Symposium* p.207-214.
45. McDonald, S. E., R. W. Tinus and C. P. P. Reid. 1984. Modification of ponderosa pine root systems in containers. *J. Environ. Hort.* 2(1):1-5.
46. McDonald, S. E., R. W. Tinus, C. P. P. Reid and S. C. Grossnickle. 1984. Effect of CuCO₃ container wall treatment and mycorrhizae fungi inoculation of growing medium on pine seedling growth and root development. *J. Environ. Hort.* 2(1):5-8.
47. Mengel, K. and E. A. Kirby. 1982. *Principles of plant nutrition.* 3rd Edition. International Potash Institute. Bern, Switzerland.
48. Moss, A. 1971. An investigation of basal sweep of lodgepole pine and shore pines in Great Britain. *Forestry* 44:43-65.
49. Nicolosi, R. T. 1981. Influence of backfill and soil density on root growth of urban plants. *New Horizons.* pp.45-51.
50. Nussbaum, J. J. 1969. Chemical pinching for roots of container plants. *Calif. Agr.* 23(10):16-18.
51. Pellet, Harold, Margaret Litzow and Laurie Mainquist. 1980. Use of metal compounds as root pruning agents. *HortScience* 15(3):308-309.

52. Perry, T. O. 1981. Tree roots-where they grow: implications and practical significance. *New Horizons*. pp.39-48.
53. Perry, T. O. 1982. The ecology of tree roots and the practical significance thereof. *J. Arboriculture* 9:197-216.
54. Reiger, Kurt. 1988. Root control bags: industry research and feedback. 21st Ann. Tn. Nursery Shortcourse Proceedings. pp.51-55.
55. Reiger, K. and C. E. Whitcomb. 1983. A root control system for growing trees in the field. *Proc. SNA Res. Conf.* 28:100-102.
56. Rogers, W. S. and G. C. Head. 1968. Factors affecting the distribution and growth of roots of perennial woody species. pp.280-295. In: W. J. Whittington(ed), *Root growth*. Plenum, New York.
57. Romero, A. M. 1985. Effect of cupric carbonate on root modification of containerized *Pinus caribaea* var. *hondurensis*. M.S. Thesis. New Mexico State University.
58. Ruehle, J. L. 1985. The effect of cupric carbonate on root morphology of containerized mycorrhizal pine seedlings. *Can. J. For. Res.* 15(3):586-592.
59. Salisbury, F. B. and C. W. Ross. 1985. *Plant physiology*. 3rd Edition. Wadsworth Publishing Co., Belmont, California.
60. Sass, J. E. 1958. *Botanical microtechnique*. Iowa State College Press, Ames, Iowa.
61. Saul, G. H. 1968. Copper safely controls roots of tubed seedlings. *Tree Planters Notes* 19(1):7-9.
62. Schmid, W. E., H. P. Haag and E. Epstein. 1965. Absorption of zinc by excised barley roots. *Plant Physiol.* 18:860-869.
63. Stecher, Paul G. (ed.). 1960. *Merck Index* 7th Edition. Merck and Co., Inc., Rahaway, New Jersey.
64. Stone, E. C., J. L. Jenkinson and S. L. Krugman. 1962. Root-regenerating potential of douglas-fir seedlings lifted at different times of the year. *For. Sci.* 8:288-297.

65. Stone, E. C. and E. A. Norberg. 1978. Container-induced root malformation and its elimination prior to planting. Proc. of the Root Form of Planted Trees Symposium. British Columbia Ministry of Forest/Canadian For. Serv. pp.65-72.
66. Struve, Daniel K. and Bruno C. Moser. Auxin effects on root regeneration of scarlet oak seedlings. J. Amer. Soc. Hort. Sci. 109:91-95.
67. Thomson, W. T. 1978. Agricultural chemicals - book IV - fungicides. Thomson Publications, Fresno, California.
68. Ticknor, R. L. 1989. Production of forsythia plants for forcing. Proc. Int. Nat. Prop. Soc. 39:115-118.
69. Tilt, Ken and Hermon Dickerson. 1987. Effects of irrigation on growth of four ornamental trees produced in grow-bags and by traditional field production methods. Proc. SNA Res. Conf. 32:156-158.
70. Tilt, Ken and Hermon Dickerson. 1987. Growth of two tree species in field containers versus conventional field production and effect of first year fertilization rates. Proc. SNA Res. Conf. 32:153-155.
71. Tilt, K., W. T. Witte and Hermon Dickerson. 1988. Effects of irrigation on growth of four ornamental trees produced in grow-bags and by traditional field production methods. Proc. SNA Res. Conf. 33:114-117.
72. Tisdale, Samuel L. and Werner L. Nelson. 1975. Soil fertility and fertilizers. 3rd Edition. McMillan Press, New York.
73. Wainwright, S. J. and H. W. Woolhouse. 1975. Physiological mechanisms of heavy metal tolerances in plants, p.231-257. In: M. J. Chadwick and G. T. Goodman. The ecology of resource degradation and renewal. Blackwell, Oxford.
74. Watson, Gary. 1985. Tree size affects root regeneration and top growth after transplanting. J. Arboriculture. 11:37-40.
75. Watson, G. W. 1986. Cultural factors can influence root development for better transplanting success. J. Environ. Hort. 4:32-34.
76. Watson, G. W. and E. B. Himelick. 1982. Root distribution of nursery trees and its relationship to transplanting success. J. Arboriculture 8:225-229.
77. Watson, G. W. and E. B. Himelick. 1982. Seasonal variation in root regeneration of transplanted trees. J. Arboriculture 8:305-310.

78. Wenny, D. L., Y. Liu, R. K. Duncan and H. L. Osborne. 1988. First year growth of chemically root pruned containerized seedlings. *New Forests*. 2:111-118.
79. Wenny, David L. and Richard L. Woollen. 1989. Chemical root pruning improves root system morphology of containerized seedlings. *WJAF* 4(1):15-17.
80. Whitcomb, Carl E. 1984. Plant production in containers. Lacebark Publications. Stillwater, Oklahoma.
81. Whitcomb, Carl E. 1987. Establishment and maintenance of landscape plants. Lacebark Publications. Stillwater, Oklahoma.
82. Witherspoon, W. R. and G. P. Lumus. 1986. Root regeneration, starch content, and root promoting activity in Tilia cordata cultivars at three different digging-planting times. *J. Environ. Hort.* 4:76-79.
83. Yadav, Uday K. and Dewayne Ingram. 1987. Observations on transplanting from field-grow containers in Florida. *Proc. SNA Res. Conf.* 32:162-163.
84. Zimmerman, Martin H, and Claud I. Brown. 1977. Tree structure and function. Springer-Verlag. New York.

VITA

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