

# **Effects of Soil Amendments and Biostimulants on the Post-transplant Growth of Landscape Trees**

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# EFFECTS OF SOIL AMENDMENTS AND BIOSTIMULANTS ON THE POST-TRANSPLANT GROWTH OF LANDSCAPE TREES

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(ABSTRACT)

Use of soil amendments at planting is one of the time-honored traditions in horticulture, although their effectiveness has been questioned by many. Recently, humate and humate-based products, generally known as biostimulants, have been marketed to increase transplant success. In this study, three experiments were conducted to examine the effects of soil amendments and biostimulants on post-transplant growth of landscape trees. The first experiment, conducted in a greenhouse, determined the effects of several biostimulant treatments (granular humate, water-soluble humate, liquid humate, liquid humate+ = humic acid, hormones, and vitamins) and fertilizer levels (low, medium, high) on the growth of container-grown *Corylus colurna* L. (Turkish hazelnut) seedlings. Biostimulants did not increase top growth compared to control treatments, but root growth was increased by granular humate at a medium fertilizer rate. The second experiment examined the effects of biostimulants (granular humate, water-soluble humate, liquid humate+) on the post-transplant root growth and sap-flow of landscape-sized balled and burlapped *Acer rubrum* L. (red maple) grown in root observation compartments (rhizotrons). Biostimulants did not increase root growth over control treatments, but sap-flow was increased. The third experiment, conducted in the field (Groseclose silt loam soil) investigated the effects of soil amendments (peat, and compost) and biostimulants (granular humate, and liquid humate+) on the post-transplant growth of *Crataegus phaenopyrum* (Blume) Hara (Washington hawthorn) and red maple transplanted bare-root, and grown under combinations of irrigated vs non-irrigated and fertilized-at-planting vs non-fertilized-at-planting regimes. Hawthorn controls generally had less top growth than the other soil treatments as a whole. No soil treatment was higher than control for top growth of red maple. However, root growth of red maple was highest in the peat-treated trees. Stem diameter and dry mass for the control and compost treatments were higher than the biostimulant treatments in irrigated plots, but no differences were observed in non-irrigated plots. Granular humate-treated trees resulted in higher stem diameter and dry mass than the liquid humate+-treated trees in non-irrigated plots. There were no effects of fertilizer, or irrigation on growth after two growing seasons for either species.

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## Chapter One

### Introduction

Landscape trees are produced in nurseries in a relatively controlled environment. Upon transplanting in the landscape, they are subjected to high levels of post-transplant stress, caused by such factors as root loss, water stress, insects and disease, and soil changes (Koller, 1977). Therefore, in the initial stages of establishment it is essential to minimize stress for plants with the best growing conditions possible.

Soil properties such as: water holding capacity, cation exchange capacity, bulk density, and soil fertility directly affect plant development. In soils where these properties are poor, conditions are often improved by the addition of soil amendments, including organic materials, synthetic chemical fertilizers, or humate-based products. Soil amendments are thought to aid plant establishment by providing for a rooting environment from the improvement of soil structure, aeration, water retention, and nutrient availability (Wagar, 1982; Corley, 1984; and Autio et. al., 1991).

Many studies involving soil amendments and plant establishment have been conducted over the past two decades with varying results; with some studies showing that soil amendments improve plant establishment (Wagar, 1982; Autio et. al. 1991; and Waller, 1994), but others have shown no significant improvement (Schulte, 1975; Whitcomb, 1979; Ingram, 1981; Corley, 1984; Hummel, 1985; Watson, 1992; Smalley and Wood, 1995). Presently, because of many conflicting results, there is no consensus on whether or not soil amendments should be applied at planting.

Application of inorganic fertilizers at planting time is another unresolved issue. Some studies indicate that the application of inorganic fertilizers aid plant establishment (Schulte and Whitcomb, 1975; and Autio et. al., 1991). On the other hand, Whitcomb (1979) found that the application of fertilizers did not enhance post-transplant growth of several landscape tree species. Additionally, one study showed that the application of inorganic fertilizer only improved plant establishment when fertilizer was applied with organic amendments (Ingram, 1981). In the few studies where inorganic fertilizers and soil amendments are used in combination, interactions between the fertilizers, amendments, and irrigation regimes were not adequately tested (Corley, 1984; and Worley, 1991).

Some researchers have reported that biostimulants can reduce plant stress as well as enhance plant nutrient uptake, and decreasing the need for inorganic fertilizer for plant growth (Berlyn and Russo, 1990). Berlyn and Russo (1990), developers of the biostimulant ROOTS™, say that ROOTS™ reduces plant water stress by increasing plant root growth after transplanting thus increasing a plant's ability to absorb water, and nutrients. Webb and Biggs (1988) noticed an increase in water absorption of 23yr old declining citrus trees after being treated with humate. However, others studies with biostimulants or their individual components have had less encouraging results (Albregts, 1988; Csizinszky, 1990; Laiche, 1991; Elliott and Prevatte, 1996).

Therefore the following study was conducted to examine the effects of soil amendments and biostimulants on tree growth under various planting regimes.

## Chapter Two

### Literature Review

#### Soil Amendments

Tree growth response to soil amendments has received considerable attention over the past two decades. The goal for the majority of soil amendment research has been to identify the best growing conditions possible for tree establishment. Most would agree that a soil with good structure, good aeration, good water holding capacity, and adequate nutrient availability would provide these conditions. In soils where these qualities are poor, soil amendments are often added to improve the planting environment.

#### I. Compost:

Tester (1990) demonstrated that soil properties and fertility of a sandy loam soil (Evesboro loamy sand soil) can be improved with the addition of organic materials. Tester conducted two studies over a five year period on the establishment of tall fescue. The first study examined changes in penetration resistance, bulk density, soil water content, N and P content, and pH of the soil after a single application of sludge compost, or the addition of a traditional chemical fertilizer. Compost improved growing conditions for the tall fescue after five years. The improvement was attributed a decreased bulk density, increased soil water retention and more nutrient availability. Grass treated with compost resulted in better establishment than grass treated with chemical fertilizers.

In a second study, Tester examined the effects yearly applications of compost and traditional chemical fertilizer on organic matter content, water retention, bulk density. Several vegetable crops were planted sequentially throughout the growing season. Compost was applied once at the beginning of each season, and chemical fertilizer was applied at the beginning of each new planting. After five years, compost increased the organic content, water holding capacity, and bulk density, whereas the chemical fertilizer treatments had less effect.

In contrast to Tester's study on grass, Watson et. al. (1993) showed that soil amendments may not be needed for good tree establishment. Watson's study, soil amendments were added to planting backfill for container-grown *Cotoneaster apiculata* L. and *Juniperus chinensis* L. 'Pfitzeriana'. Treatments included: no backfill = unamended site soil; 25% mushroom compost and backfill; and 40% sand, 10% leaf compost and backfill. Root density and shoot growth were evaluated after 14 months. There were no treatment differences on root density or shoot growth.

Hodge (1995) studied the effects of seven organic amendments on backfill soil characteristics and *Quercus robur* L. in clayey, chalky, and silty type soils. After three growing seasons, three of the amendments resulted in tree survival ratings that were higher than controls at the clay site exclusively. High soil electrical conductivity was found to effect tree survival the most after the first growing season. Hodge attributed the high conductivity for some of the amendments to lack of standardization in manufacturing. Concentrations of ammonia, phosphorus, and potassium, high enough to be detrimental to tree growth, were found in several

of the amendments. Hodge concluded that soil amendments may not be needed in disturbed native soils and suggested that disturbance of the soil from the planting process may be sufficient for good establishment.

## II. Humate:

Humic substances are organic compounds that result from the decomposition of plant and animal material. Soil organic matter is composed of 0 to nearly 100 percent humic acids (Schnitzer, 1967). These acids are important constituents of soils in that they affect water retention, contribute to cation exchange capacity, and serve as a nutrient reserve for living organisms, plants and microbes. The ability of humic acids to act as a nutrient reserve comes from having a high exchange capacity, and the capability to form water soluble complexes with metal ions ,e.g. Fe, thus possibly enhancing the absorption of some ions by roots (Schnitzer, 1967). Additionally, humic substances are said to behave similar to auxins, and promote root growth of some plants (O'Donnell, 1973). Humic substances are called acids when hydrogen ions dominate exchange sites located on the humic molecule. When these hydrogen ions are replaced by other cations the humic structure is referred to as humate (Senn and Kingman, 1975; and Webb and Bings, 1988). Humate has been the focus of many studies in past years.

Webb and Bings (1988) examined the effect of humate on stressed citrus trees. A combination of humate and micronutrients, or humate and  $\text{CaNO}_3$  increased fruit set, growth flushes, bark thickness, and production on 23yr old *Citrus reticulata* L. `Honey Tangerine` trees that previously showed signs of stress. The affects of various humate amounts (920g, 1846g, or 2760g), incorporated directly into soil, on water uptake of 23yr old *Citrus sinensis* L. `Valencia` sweet orange on rough lemon root stock were also studied. Humate-treated plants had a slight increase in water uptake values the first 12 months of the study compared to control plants (no humate). Webb and Bigg, treated `Hamlin` sweet orange on `Cleo` rootstock and `Ruby Red` grapefruit on `Swingle` with 0g, 230g, 460g, 920g, or 1846g of humate at planting. Cross sectional trunk area increases were measured, and revealed that the humate-treated trees had a greater increase in stem size compared with controls over a 10-11 month period.

Humate has also been shown to improve growth of `Chardonnay` grapevines, applied as the commercial humate product "Gro-Mate"(GM) (Reynolds et. al., 1995). Growth measurements were determined after treatment with 5 different rates of granular GM (0,8,16,32, or 64g/pot), and two different liquid applications of the humate (1x or 2x/week). Optimum humate rate for shoot extension was 32g/pot. Liquid treatments decreased all aspects measured. The higher granular GM rates (64g/pot), and the liquid humate applications caused chlorosis and necrosis, and cupping of leaves. The authors theorized that root damage resulting from high salt concentrations may have led to these foliar characteristics. Nutrient analysis of the petioles and lamina, and nutrient analysis of the planting soil, revealed toxic levels of some essential nutrients. Reynolds et. al. concluded that humate products are beneficial to plant growth when used at moderate levels.

In contrast to the literature on humate just discussed, a study by Laiche (1991) demonstrated that humic acid can have a negative effect on growth of some container-grown

landscape plants. Laiche evaluated the effectiveness of humic acid and slow release fertilizers on the growth index, fresh weight, and root rating of four common landscape plants: *Ilex crenata* L. 'Compacta'; *Ilex vomitoria* L. Straughn's selection; *Rhododendron* L. 'Pride of Mobile'; and *Juniperus chinensis* L. 'Nick's Compacta'. When humic acid was used alone at increasing rates, the growth index and fresh weight of *Ilex crenata* and *Ilex vomitoria* was decreased; and the growth index of *Juniperus chinensis* was reduced. Humic acid was also applied together with two different rates of fertilizer (805g, or 1380g/.76m<sup>3</sup> of 16-6-10 slow release fertilizer) and four rates of humic acid (0g, 460g, 690g, or 920g/.76m<sup>3</sup>). After eight months, the humic acid/fertilizer combination did not effect growth, except for increased fresh weight of *Ilex crenata* grown at the higher fertilizer level. There were no explanations for these findings, but Laiche concluded that humic acids may be detrimental to growth of woody plants.

### III. Biostimulant products:

In addition to humates, biostimulants are said to promote plant growth, and are being used increasingly in ornamental horticulture. Biostimulants are defined by Berlyn and Russo (1990) as being, "non-fertilizers which benefit plant growth," containing natural products such as cytokinins and humic acids often obtained from seaweed extracts. These extracts have been shown to improve the growth of some plants (Crouch et. al., 1992). Biostimulant products are said to increase a plant's nutrient and water uptake, resistance to water stress, and resistance to residual herbicides in the soil (Schmidt,1990; Russo and Berlyn ,1990). Studies with trees, and bedding plants have shown that use of biostimulants can improve plant growth. For example, Russo et. al.(1990), in their studies with the biostimulant Roots<sup>TM</sup> (a root dip containing a mixture of seaweed, humic acid and vitamins), demonstrated that several nursery grown trees (loblollypine, red maple, and sand pine) can benefit when Roots<sup>TM</sup> is used as a root dip prior to planting in the field. They found increased root and shoot growth, growth potential, and stress resistance, attributing these effects of Roots<sup>TM</sup> to increased chlorophyll content (demonstrated in same study with rye grass), and root regeneration capacity (demonstrated with Black Walnut and Bentgrass).

Roots<sup>TM</sup> has also been shown to improve the growth of bedding plants when used as a soil drench. Poincelot (1993) found that by drenching the planting media with a Roots<sup>TM</sup> solution prior to sowing geranium seeds (cultivar 'Scarlet Border'), germination rates and development were improved compared to controls (no Roots<sup>TM</sup>). Poincelot also found improved root and shoot production by soaking the seedlings (ones that received Roots<sup>TM</sup> as a soil drench before seed sowing) in a root dip solution at transplanting.

Studies with biostimulants have not always shown improved plant growth. For example, a study by Csizinszky (1990), using two cultivars of bell peppers ('Early Calwonder', and 'Jupiter'), demonstrated that biostimulants had no influence on yields or nutrient content of the peppers. Six granular and liquid biostimulants were applied according to the manufacturer's recommendations. Even though some of the biostimulants increased the fruit yield in earlier harvests, marketable yields of all biostimulant treatments were not different. 'Early Calwonder' peppers treated with biostimulants produced similar fruit yields as control treatments; but for 'Jupiter' peppers treated

with biostimulants, fruit yields were less than controls. Nutrient levels for 'Early Calwonder' pepper were at or above those needed plant growth for all treatments. Even though some of the biostimulant treatments increased levels of some nutrients, these increases did not have any effect on the marketable yield of the peppers. Higher-than-normal yields were attributed to differences in the production system and cultivar used, fertilizer treatments, and number of harvests; rather than biostimulants. However, Csinzinszky states that because of their cytokinin content, biostimulants may be beneficial in periods of plant stress. According to his theory, internal plant production of cytokinin may be limited during periods of plant stress, and cytokinin contained in biostimulants may be beneficial.

In another study evaluating the benefits of biostimulants on plant growth, Albrechts et. al., (1988) demonstrated the effect of a number of biostimulants (foliar sprays of various compositions) on the fruit yield and weight of several strawberry clones ('Chandler'; 'Dover'; 'Pajaro'; and Florida breeding line 79-1126). A rate study with humic acid was also conducted on the 'Chandler' and 'Dover' clones. Biostimulant rates and time of application varied with brand of biostimulant used, and the humic acid rates ranged from 0 to 1kg/24m<sup>2</sup>. Biostimulant and humic acid treatments did not increase yield or fruit weight compared to controls after three growing seasons. Albrechts et. al. speculate that because of a cover crop used in conjunction with strawberry planting, benefits of the biostimulant or humic acid treatments may have been reduced by the constant level of soil organic matter maintained by the cover crop. The authors concluded that if proper planting techniques (e.g. cover crop) are followed, the use of biostimulants is unwarranted.

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## Chapter Three

### Effect of Biostimulants on the Growth of Container-grown *Corylus colurna* L. (Turkish hazelnut) Seedlings Grown Under Greenhouse Conditions

#### Introduction

Biostimulants have been described as “non-nutritional products that may reduce fertilizer use and increase yield and resistance to water and temperature stresses ” (Russo and Berlyn, 1992). Major components of commercially available biostimulants include: humates (granular or liquid forms), plant growth hormones (e.g. cytokinins), and various metabolites. These components, or combinations of components, have been shown to increase plant shoot and root growth, and uptake of some nutrients (Tan and Nopamonbodi, 1979; Russo and Berlyn, 1992; Sanders et. al., 1990; and Poincelot, 1993).

Russo and Berlyn (1990, and 1992) found that the growth of several tree species, turfgrasses, and vegetable crops is improved with a biostimulant called Roots™, which consists of humic acid, cytokinin, thiamine, and ascorbate. Likewise, Poincelot (1993) observed the beneficial effects of Roots™ on the growth of tomato, broccoli, and several flowering annuals. However, other research using Roots™ found no advantage to growth and yield of cabbage plants (Heckman, 1995). Additional studies with biostimulants similar to Roots™ have found no advantages in their use to promote plant growth (Albregts et. al., 1988; Elliott and Prevatte, 1996).

The effect of humic acid, a major component of biostimulants, on plant growth has been studied extensively (O'Donnell, 1973; Vaughan, 1974; Lee and Bartlett, 1976; Rauthan and Schnitzer, 1981; Laiche, 1991; Tattini et. al., 1991; and Reynolds et. al. 1995). A number of researchers have found that humic-like substances can increase root growth (Schnitzer and Poapst, 1967; O'Donnell, 1973; Vaughan, 1974; Mylonas and McCant, 1980). Vaughan (1974) proposed that humic acids may primarily increase root growth by increasing root cell elongation. Others have found that humic-like substances can produce root systems with increased branching and numbers of fine roots, and as a result potentially increase nutrient uptake by way of more root surface area (Rauthan and Schnitzer, 1981).

Few studies have involved the effects of biostimulants on growth of container-grown trees. Therefore, the purpose of this study was to determine the effects of several biostimulant type products on the overall growth of Turkish hazelnut transplanted bare-root to containers and grown under greenhouse conditions. Turkish hazelnut was chosen because its relatively unbranched root system (Harris et. al., 1994) makes it a good model to test possible effects of biostimulants on root system morphology. In addition, the interactive effect between biostimulants and different fertilizer rates was examined.

## Materials and Methods

This experiment was conducted at the Virginia Tech greenhouse complex in Blacksburg, VA from 28 February, 1996 to 17 June, 1996. One-hundred-and-fifty bare-root *Corylus colurna* L. (Turkish hazelnut) seedlings from Heritage Seedlings, Inc. Salem, OR (mean height = 33.8cm, and mean stem diameter = 5.8mm) were planted in 3.7L plastic pots in a pine bark substrate, amended with 4.0 kg dolomite, and 1kg Micromax™ (The Scotts Co., Maryville, OH) per m<sup>3</sup>. After planting, seedlings were pruned to a central leader, and initial height and stem diameter were measured. Plants were watered as needed throughout the experiment so as to assure a drought-free condition.

The following soil treatments were applied at planting: 1) control- amended pine bark substrate only; 2) granular humate; 3) wetttable soluble humate; 4) liquid humate; and 5) liquid humate +. Granular humate (Earthgreen Products, Dallas, TX) was applied at 2g/container. Wetttable soluble humate (Menefee WSP™, Earthgreen Products, Dallas, TX) was applied as a soil drench after planting at 2.5mg dissolved in 180ml of H<sub>2</sub>O and applied to each replication. Liquid humate (Growplex™, Earthgreen Products, Dallas, TX) was applied as a 30 minute root soak (30ml Growplex™ mixed with 4L of H<sub>2</sub>O) immediately prior to planting. Liquid humate + (Roots™, Roots Inc., New Haven, CT) was applied as a 30 minute root soak (20ml Roots™ in 1L of H<sub>2</sub>O) immediately prior to planting. All treatments were applied at the manufacture's recommended rate.

To examine interaction effects between the soil treatments and fertilizer rates, a top-dressing of slow release fertilizer (Osmocote: 18N-2.6P-9.9K, The Scotts Company) was applied at either 1.62, 2.52, or 3.42g of N per container. These are the manufacturer's recommendations for low, medium, and high rates, respectively. There were 10 single plant replications of each soil treatment x fertilizer combination.

At the conclusion of the experiment, final height and stem diameter measurements (5cm from substrate) were recorded, and leaf and stem samples were dried to a constant mass at 70<sup>0</sup> C and weighed. For root length analysis, one root system per treatment, at the medium fertilizer rate, was randomly selected. Each root system was separated into five diameter classes (0-1mm, 1-3mm, 3-5mm, 5-10mm, and >10mm), and subsequently divided into subsamples of varying amounts for determination of length-dry mass relationships. Root length was determined using a combination of computer imaging software (Desk-Scan II, Hewlett Packard Co.), and a computer-image analyzing system (Delta-T SCAN, Delta-T Devices LTD. Cambridge, England). Subsamples were then dried to a constant mass at 70<sup>0</sup> C and weighed. A relationship between dry weight and root length for each diameter class was made using a simple-linear regression procedure (SAS vers. 6.08, Cary N.C.) from which regression equations were generated for each root diameter class.

Four replications from each treatment were randomly chosen from the remaining root systems. Each root system was separated into five root diameter classes, and dry mass was determined as described earlier. Root length for each diameter class was determined for each replication using the previously defined dry mass:length relationship.

The experiment was a completely random design. Height, caliper, and length data were analyzed using the analysis of variance procedure followed by Tukey's HSD to separate mean values (SAS vers. 6.08, Cary, N.C.).

## **Results and Discussion**

*Top growth.* Top growth of biostimulant-treated trees was the same as the controls except for liquid humate and liquid humate + treated trees which resulted in the lowest height growth. The liquid humate + had the greatest negative effect on stem diameter growth, and leaf and shoot dry mass. Higher fertilizer rates had the highest stem diameter growth, and leaf and shoot dry mass. Many others have demonstrated that as fertilizer rate is increased, top growth will increase, (e.g. Yeager and Wright, 1981) (data not shown). Finally, there were no significant interactive effects of soil treatments x fertilizer rates for any top growth parameter (Table 1).

These results confirm findings of others who have observed the inconsistent effects of biostimulants on plant top growth. For instance, Albregts et. al. (1988) found no increase in the fruiting of strawberry after application of several types of biostimulants. Heckman (1995) demonstrated that the yield of cabbage was not increased over controls with biostimulants. Elliot and Prevatte (1996) showed that seaweed-based biostimulants did not improve turfgrass growth. Laiche (1991) found that the use of humic acid (a major component of some biostimulants) can be detrimental to the growth of several container-grown woody plant species.

Additionally, results from Table 1 suggest that use of liquid humates, as applied in this study, may have a negative impact on plant growth. Reynolds et. al. (1995) showed that the application of liquid humate reduced the top growth of 'Chardonnay' grapevines; but the application of a granular form of the same product had a positive effect on grapevine growth. In the present study, granular humates performed better than the liquid humates. However, as previously stated, results of the granular humate treatments were not different than controls for any aspect of top growth.

In this study, there were no significant interactions between fertilizer rates and biostimulant treatments for any parameter of top growth which was in agreement with other studies using container-grown plants. For example, Laiche (1991) found that plant growth generally was not increased by humic acid treatments in conjunction with fertilizer treatments compared to plants that just received fertilizer. Similar findings were seen by Heckman (1995) study with cabbage, and by Albregt et. al. (1988) using strawberry.

*Root length.* The majority of root length occurred within the 0-1mm diameter class, where liquid humate and liquid humate + treatments resulted in the lowest lengths (Table 2). Reynolds et. al. (1995) found that liquid-applied humates reduced root dry weights of grapevines as compared to granular humates which increased root dry weights, and concluded that root damage may have occurred because of high salt concentrations produced by liquid humate treatments. Similarly, in the present study, granular humates had higher root lengths compared to liquid humates in the 0-1mm diameter class; however, salt levels of liquid humates were within

tolerable levels for trees grown in containers (.6dS/m) (Niemiera, personal communication). Additionally, pH levels for liquid and granular humate soil solutions averaged 5.8 and 5.9, respectively. Fine roots are generally expected as having the greatest absorption capacity for water and nutrients as compared to thicker diameter roots. Therefore, further investigation is needed to examine why liquid applied humates had such detrimental effects on fine root growth in this study.

With the exception of the 3-5mm diameter class, fertilizer treatments did not have an effect on root length (Table 2). The interactive effects between soil treatments and fertilizer rates were significant for the 5-10mm diameter class (figure not shown) and total root length (Figure 1). The effect of fertilizer treatments on the 3-5mm diameter class, and the interaction for 5-10mm diameter class do not effect root length substantially, since in this case, these diameter classes represent small percentages of total root length (Table 2). Figure 1 illustrates the interactive effect of fertilizer x biostimulant treatments on total root length. Granular humate increased growth at the 2.52g N/container rate. Some researchers have hypothesized that humate-like substances can increase plant nutrient uptake. Rauthan and Schnitzer (1981), studying the effect of fulvic acid on growth of corn, found that nitrogen content and dry weight of corn roots were increased by moderate rates (100-300 ppm) of fulvic acid. Several theories were stated to explain this effect of fulvic acid: 1) increased root cell membrane permeability; 2) increased surface area of roots by causing the formation of more root hairs and root branching, thus making nutrient absorption more efficient; and 3) may act as a plant hormone and aid translocation of nutrients throughout the plant. However, in the current study, the reason that a increase of root length on granular humate-treated plants occurred at the medium fertilizer rate and not low and high rates is unknown. More investigation is needed to explain this effect.

In this study no beneficial effects were observed on tree top growth with the use of biostimulants. Furthermore, use of liquid humates at the rates used in this study, had a negative effect on root and shoot growth of container-grown trees. However, total root length of granular humate-treated trees was higher at the medium (2.52g N/container) fertilizer rate. Increased root length relative to top growth is generally thought to produce a plant with greater potential for post-transplant growth. Results of this study should be confirmed with other species, and post-transplant studies implemented, before general recommendations are made.

Table 3.1 Effects of biostimulant and fertilizer treatments on height growth (increase), stem diameter growth (increase), and leaf/shoot dry mass of Turkish hazelnut.

Treatment	Height growth (cm)	Stem diameter growth (mm)	Leaf dry mass (g)	Shoot dry mass (g)
Control	37.2a <sup>Z</sup>	3.0a	8.3a	10.5a
Granular humate	35.8a	2.8a	7.9a	10.1a
Wettable soluble humate	33.6a	2.6a	7.6a	10.0a
Liquid humate	20.1b	1.7b	5.0b	7.1b
Liquid humate +	8.9b	0.8c	2.2c	4.8c
Treatment	0.0001 <sup>Y</sup>	0.0001	0.0001	0.0001
Fertilizer	0.1258	0.0052	0.0146	0.0142
T x F	0.2284	0.1685	0.1756	0.1988

<sup>Z</sup>Means in columns followed by the same letter are not significantly different, Tukey's HSD ( $P \leq 0.05$ ,  $n = 30$ ).

<sup>Y</sup> $P > F$

Table 3.2 Effects of biostimulant and fertilizer treatments on root length (m) of Turkish hazelnut for four different diameter classes, and total root length.<sup>Z</sup>

Treatment	Diameter Class				Total (m)
	0-1mm	1-3mm	3-5mm	5-10mm	
Control	176.1ab <sup>Y</sup>	11.7a	1.3a	0.8a	189.9
Granular humate	216.3a	3.3b	0.6b	0.01b	220.3
Wettable soluble humate	158.4b	9.6a	0.9ab	0.7a	169.7
Liquid humate	53.5c	3.9b	1.1a	0.8a	59.4
Liquid humate +	24.1c	1.9b	0.6b	0.2b	26.8
Treatment	0.0001 <sup>X</sup>	0.0001	0.0002	0.0001	0.0001
Fertilizer	0.8673	0.6982	0.0024	0.7837	0.8598
T x F	0.0601	0.3546	0.1613	0.0028	0.0509

<sup>Y</sup>Means in columns followed by the same letter are not significantly different, Tukey's HSD ( $P \leq 0.05$ ,  $n=12$ ).

<sup>X</sup>  $P > F$

<sup>Z</sup> Roots >10mm in diameter excluded from table;  $n=12$ .

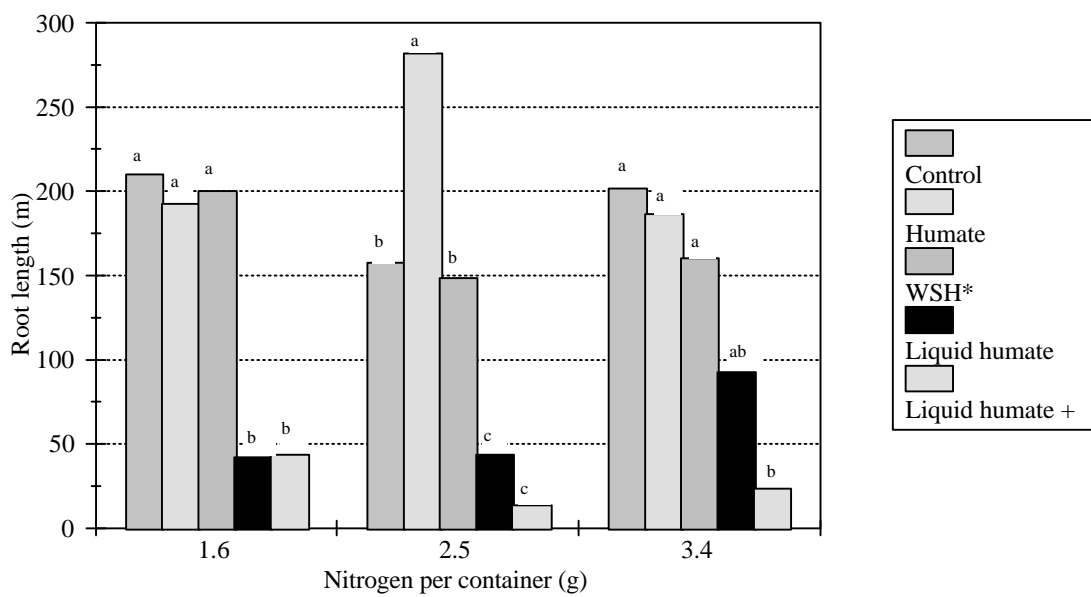


Figure 3.1 Effects of fertilizer and biostimulant treatments on total root length on container-grown Turkish hazelnut. Means with the same letter are not significantly different, Tukey's HSD ( $P = 0.05$ ,  $n = 4$ ).

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## Chapter Four

### Effect of Biostimulants on Early Post-transplant Root Growth and Sapflow of Balled and Burlapped Red Maple

#### Introduction

Reduced post-transplant growth (transplant shock) of field-grown trees is thought to be primarily a factor of plant water stress caused by the loss of root density resulting from the transplanting process (Haase and Rose, 1993). As little as 2% of the original tree root system is retained upon transplanting (Watson and Himelick, 1983). Consequently, the root system's capacity to absorb nutrients and water is greatly reduced. Therefore, rapid root regeneration is vital for restoration of full nutrient and water absorption capability, thereby increasing the chance for post-transplant survival.

Many researchers have found that tree root growth can be enhanced with the use of plant growth hormones (e.g. auxins) applied at planting (Hartwig and Larson, 1980; Prager and Lumis, 1983; Kelly and Moser, 1983; and Struve and Arnold, 1986). For example, Lumis (1982) found that the root growth of landscape-size red oak trees can be enhanced by the application of IBA (indolebutyric acid) to the root systems before planting.

Recently, new growth-promoting products called biostimulants have been developed, and have been shown to improve root growth of a number of plant species (Russo and Berlyn, 1990; Sanders et. al., 1990; Poincelot, 1993; and Reynolds et. al., 1995). These products often consist of organic compounds (e.g. humates or seaweed extracts), as well as growth hormones (e.g. cytokinins). Improved plant growth from application of biostimulant products containing seaweed extracts may be a result of auxins and cytokinins (Crouch et. al., 1992; and Tay et. al., 1987). Furthermore, humic substances are claimed to increase root growth of various plant species in a manner similar to auxins (O'Donnell, 1973; Tattini et. al., 1991). However, not all research has shown biostimulants to be beneficial to root growth (Laiche, 1991; Elliott and Prevatte, 1996).

There is some evidence that humic products may increase the nutrient and water uptake by plant roots, Russo and Berlyn (1990) propose that some biostimulants may increase water uptake of some plants. Similarly, Webb and Briggs (1988) noted that water uptake of declining citrus trees was increased when humates were applied. This enhanced uptake of water and nutrients may be an effect of increased root surface area or increased cell permeability caused by the humates (Rauthan and M. Schnitzer, 1981). The purpose of this study was to examine the effects of several biostimulants on root regeneration and water uptake of landscape-sized *Acer rubrum* L. 'Red Sunset' (Red maple) moved with intact root balls (balled and burlapped), and transplanted into root observation compartments (rhizotrons).

## Materials and Methods

This experiment was conducted at the Virginia Tech Urban Horticulture Center, Blacksburg, VA (USDA hardiness zone 6a). Sixteen *Acer rubrum* L. 'Red Sunset' (Red maples) were hand dug on 12 April, 1996 from a Groseclose silt loam (clayey, mixed, mesic Typic Hapludults) field soil with pH 6.2. Trees were moved with intact root balls (35cm in diameter) wrapped in burlap (ball and burlapped = B&B) according to American Association of Nurserymen (1990), and transplanted into single tree rhizotrons (described below) which allowed for continuous observation of root growth. The planting substrate was a sand-peat mixture (50:50 v:v), and all trees were fertilized at planting with Osmocote (18N-2.6P-9.9K, The Scotts Co.) at a rate of 19.4g N per tree. Mean height and stem diameter (10cm above soil) at time of planting were 2.5m and 24mm respectively. Throughout the experiment, trees were irrigated using a micro-irrigation system so as to maintain plants near field capacity.

Treatments were applied on 16 April, 1996, as: 1) control - no soil amendment; 2) liquid humate; 3) liquid humate +; and 4) granular humate. The liquid humate (Menefee Humate WSP™, Earthgreen Products, Dallas, TX) was applied as a soil drench after planting at a rate of 14 mg dissolved in 1 L of H<sub>2</sub>O, and each replication received 2.26 L of the solution. The liquid humate + (Roots™, Roots Inc., New Haven, CT) was applied as a soil drench after planting at 90 ml per 11.31 L and applied to each replication. The granular humate (Earthgreen Products, Dallas, TX) was applied as a top dressing at 6.3g per rhizotron. All treatments were replicated four times, and applied at the manufacturer's recommended rate. The experimental design was completely random with 4 single tree replications per treatment.

Rhizotrons (one rhizotron per replication) were constructed from Keeper-Uppers (KU) (Mobile, AL), normally used to protect trees from possible windthrow and container temperature changes in an above-ground containerized production system. KUs are flat-topped shells, shaped like pyramids with a square base (53cm length x 54cm width x 38cm height), and an opening (43cm in diameter) in the top to the ground below for plant-growing containers (Harris et. al., 1996). These rhizotrons were painted white, and insulated using reflective plastic bubble insulation (Reflectix Insulation, Reflectix Inc., Markleville, IN) to maintain a planting substrate temperature comparable to the native soil. The rhizotron and native soil temperatures were monitored using thermocouples. The average daily temperature throughout the experiment was 19.5C for the rhizotrons and adjacent native soil.

Tree roots were observed through polycarbonate plate windows (28cm x 28cm, Lexan®XL, GE Worldwide Manufacturing Sites, Mount Vernon, IN) fashioned to the side of each rhizotron. Windows were placed on the south side of each rhizotron, and a 5cm x 5cm grid was permanently marked on each plate from which root length was determined using the line intersect method first developed by Newman (1966), then modified by Marsh (1971) and Tennant (1975) who established the relationship that:

$$\text{Root Length} = 11/14 \times \text{number of intersections} \times \text{grid size}$$

Each pane was covered with a 5cm thick styrofoam door that served to block sun-light and insulate against temperature extremes.

Root growth was recorded for each replication weekly by taking photographs (35mm slides) of the root systems, beginning when the roots were first detected in the rhizotron windows on 20 May, 1996 and ending 29 August, 1996. To record and analyze the number of root-line intersections, slides were scanned using a 35mm slide scanner (Scanmaker 35t, Microtek International Inc., Taiwan, R.O.C.) and computer imaging software (Adobe Photoshop vers. 3.0, Adobe Systems Inc., Mountain View, CA). Intersections were read in SigmaScan software (Jandel Scientific, San Rafael, CA).

At the conclusion of the experiment on 26 Sept. 1996, all new roots visible outside the original root ball were harvested. Root dry masses were determined by drying to a constant mass at 70<sup>o</sup> C.

Two trees per treatment were randomly selected and water uptake determined by stem sapflow measurements monitored at three different periods after transplanting: 1) 2 May, 1996 to 15 May, 1996; 2) 10 June, 1996 to 16 June, 1996; and 3) 27 June, 1996 to 10 July, 1996. Sapflow was measured with a Flow32-AO Sap Flow Measurement System (Dynamax, Houston, TX) that functions under the heat balance theory for measuring plant sapflow where “ an insulated section of stem about as long as its diameter is heated externally at a constant rate, and the heat fluxes in the radial and vertical direction are measured. Heat carried in the moving sap is calculated by subtracting measured conductive heat fluxes from the heat input... ” (Steinberg, et. al. 1989). With the Flow32-AO system, a gauge (with heater) is placed around the stem of a tree, and the mass flow of sap (g/s) is calculated by a measurement of the heat required to maintain the sap at a constant temperature where the gauge is fastened, and by the temperature gradient values above and below the gauge. In this experiment, one 16mm SGB-16 gauge (Dynamax, Houston, TX) was fitted to the main trunk of each replication. Sapflow gauges were sealed with silicone for protection against moisture. Heat from the sun was minimized by an insulated collar and heat shield placed around the gauge, as well as with aluminum foil that covered the tree trunk from the gauge to the ground. Data were collected using a DNX10 Datalogger (Dynamax, Houston, TX) that was programmed to measure the sapflow rate every 60 seconds and average the values every 15 minutes. Output was recorded every 30 minutes, and downloaded to a lap-top computer for future analysis.

Root length data were analyzed by repeated measures multivariate analysis of variance, as well as single degree of freedom contrasts, and plotted over time. Final root length observed in the rhizotron windows, root dry weight, and sapflow data were analyzed using one-way ANOVA procedures. Differences among means were determined by Tukey’s HSD (SAS version 6.08, SAS Institute, Cary, NC).

## **Results and Discussion**

*Root Length.* Tree roots were first observed in the rhizotrons on 20 May, 1996, 38 days after planting. Data plotted over time for root growth can be described as first exponential until approximately 10 July, 1996, and linear thereafter for all treatments (Figure 1). Data were

therefore partitioned into these two groups and each period was analyzed separately by repeated measures.

Repeated measures analysis revealed that the effects of treatments on root length was significant over time for both periods described above (Table 1a,b), as were all contrasts performed, with the exception of control vs humate for the 20 July, 1996 thru 28 August, 1996 period (Table 1b). Between 20 May, 1996 and 10 July, 1996, root length of control and granular humate treatments was higher compared to other treatments with control and humate treatments being slightly different from each other (Figure 1, Table 1a). After 10 July, 1996 control and humate treatments maintained a similar root length, and again were higher than the other two treatments (Figure 1, Table 1b).

At the conclusion of the experiment, the granular humate treatment resulted in the highest final root length among biostimulant treatments, but this root length was the same as for the control treatment (Table 2). The liquid humate + and liquid humate treatments were also the same as compared to controls (Table 2). There were no apparent differences between biostimulant treatments for root dry mass (Table 2). However, contrary to final root length, the control treatment resulted in the lowest dry mass compared to the other treatments. This difference may be an indication that the control treatment had more fine roots, which would weigh less than thicker roots, than the other treatments.

Contrary to past studies involving biostimulants or their individual components (Schnitzer and Poapst, 1967; O'Donnell, 1973; Mylonas and McCants, 1980; Russo and Berlyn, 1990; Tattini et. al., 1991; and Poincelot, 1993), this study found little benefit in their use to promote root growth after transplanting of red maple. The granular humate treatment appeared to have a higher rate of root growth initially, but no treatment was higher than the control for root length at the conclusion of the experiment.

Other researchers have found that liquid applied biostimulants may occasionally have small effects on root growth (Reynolds et. al., 1995; Elliott and Prevatte, 1996). Reynolds et. al. (1995), using 'Chardonney' grapevines and humates, used liquid and granular humate applications of the same product, and found that liquid-applied treatments resulted in poor overall plant growth, while granular treatments resulted in improved growth. Likewise, the authors of the current study had similar results in a greenhouse experiment involving Turkish hazelnut trees treated with granular and liquid type humates (Chapter 3).

*Sapflow.* For date 1, sapflow rates for liquid humate + and granular humate treatments were highest, but only liquid humate+-treated trees had higher flow rates compared to controls. Liquid humate had the lowest flow rates which were comparable to controls (Table 2). The sapflow rates of biostimulant-treated trees were higher than controls for both dates 2 and 3. Granular humate-treated trees resulted in the highest flow rates for date 2 (Table 2). However, these rates were not different than liquid humate treatments. Likewise, liquid humate and liquid humate+ treatments had similar flow rates for date 2. There were no differences in flow rates between any biostimulant treatment for date 3.

These results support other researchers who have reported that biostimulants or their components may have a positive effect on sapflow (Webb and Bigg, 1988; Russo and Berlyn, 1990). As stated earlier, some researchers have proposed that humic substances may increase root length or root cell membrane permeability (Rauche and Schnitzer, 1981), therefore water uptake by plant roots may be increased. In the present study, root length did not appear to be increased by biostimulant treatments, although sapflow was increased. A possible explanation for the observed increase in sapflow may be an increased cell membrane permeability resulting from biostimulant treatments. However, further research is needed to examine the mechanism or mechanisms by which biostimulants might influence water uptake by roots.

In conclusion, under these growing conditions, biostimulants do not appear to increase root growth of B& B red maple after transplanting. However, the granular humate treatment may have a greater effect on root growth at another fertilizer level as was seen in Chapter 1. Biostimulants may increase sapflow soon after transplanting. How this may effect a tree's survival in the long-term has yet to be determined. In this experiment, the trees did not appear to be different visually after 20 weeks of growth, and there were no differences among treatment means for either the stem height or stem diameter growth (data not shown). However, trees in our experiment were well watered and no drought treatments imposed. Further experimentation is needed to examine biostimulant influence on tree survival in various post-transplant conditions such as drought.

Table 4.1 Repeated measures ANOVA and single degree of freedom contrasts for the effect of treatments on root length of red maple for time periods 20 May, 1996 thru 10 July, 1996, and 20 July, 1996 thru 28 Aug. 1996.

A. Root length for 20 May, 1996 thru 10 July, 1996.

Source	P > F
treatments	0.008 <sup>z</sup>
control vs others	0.005
control vs humate	0.05
control vs soil drench	0.003

B. Root length for 20 July, 1996 thru 28 Aug. 1996.

Source	P > F
treatments	0.01
control vs others	0.02
control vs humate	0.20
control vs soil drench	0.008

<sup>z</sup> P-values from Wilks' Lambda multivariate tests.

Table 4.2 Effects of treatments on final root length, root dry mass, and sapflow of red maple.

Treatments	Final root length (m)	Root dry mass (g)	Sapflow (g/day)		
			Date 1 <sup>Y</sup>	Date 2 <sup>X</sup>	Date 3 <sup>W</sup>
Control	12.1ab <sup>Z</sup>	94.5	11.3bc	21.2c	40.0b
Liquid humate	8.7b	127.2	7.6c	42.0ab	58.2a
Liquid humate+	8.8b	120.0	18.5a	33.4b	58.2a
Granular humate	12.8a	133.2	5.0ab	44.8a	58.0a
Significance	0.01 <sup>V</sup>	0.68	0.0001	0.0001	0.0002

<sup>Z</sup>means followed by the same letter within columns are not significantly different ( $P \leq 0.05$ ,  $n = 4$ ).

<sup>Y</sup>data taken 2 May, 1996 to 15 May, 1996

<sup>X</sup>data taken 10 June, 1996 to 16 June, 1996

<sup>W</sup>data taken 27 June, 1996 to 10 July, 1996

<sup>V</sup>  $P > F$

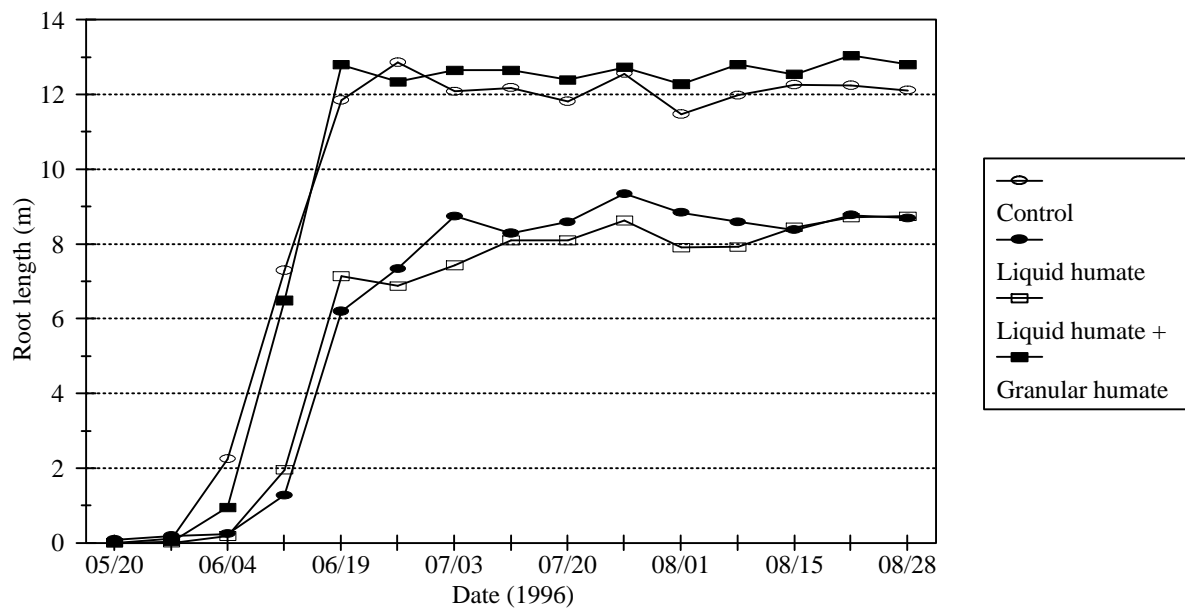


Figure 4.1 Effects of biostimulant treatments on root length of red maple transplanted into rhizotrons on 12 April, 1996. Data analyzed with repeated measures multivariate analysis, and single degree of freedom contrasts (Table 1).

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## Chapter Five

### Effects of Soil Amendments and Biostimulants on Post-transplant Growth of Washington Hawthorn and Red Maple Seedlings

#### Introduction

Trees are subjected to severe stress during the transplanting process since much of their root systems are removed upon lifting for transplant. Consequently, the capacity to absorb water and nutrients is been reduced. Therefore, for good post-transplant growth, it is important to provide plants with the best rooting environment possible.

Traditionally, soil amendments have been used to improve soil physical and chemical properties for tree growth. However, the efficacy of soil amendments on tree establishment in the landscape has been questioned over the years (Whitcomb 1979; Corley 1984; Hummel and Johnson, 1985). For example, Smalley and Wood (1995) found that several types of organic amendments had no effect on the overall growth of landscape-sized red maple trees, and concluded that the native soil available at the planting site may be sufficient for good establishment. Others believe that simply disturbing the planting site soil is the best management practice for good post-transplant growth (Hodge, 1995). Still, there is reliable research showing that treating the soil with amendments will be beneficial to plant growth in conditions where soil properties are poor (Schoenholtz et. al., 1992; Day, 1995).

Biostimulants, relatively new types of soil treatments, have recently become commercially available. Biostimulants generally consist of organic material (e.g. humates), and plant growth hormones (e.g. cytokinins), often obtained from seaweed extracts. Biostimulants may promote plant growth by increasing plant nutrient uptake, providing plant growth hormones, and acting as chelating agents for some plant nutrients, e.g. iron. They have been shown to increase overall plant growth of a number of horticultural crops and turf grasses, as well as the growth of some tree species (Russo and Berlyn, 1990; Sanders et. al., 1990; Poincelot, 1993). However not all research has shown biostimulants to be beneficial to plant growth. For example, Elliott and Prevatte (1996) found a seaweed-based biostimulant was not successful in increasing the quality or the root growth of 'Tifdwarf' Bermuda-grass. Additionally, Laiche (1991) found that humic acid (a major component of some biostimulants) applied to several different container-grown landscape shrub species can result in reduced plant growth.

Little information is available concerning how irrigation and fertilizer practices may interact with soil amendment and biostimulant treatments. Therefore, the purpose of this study was to test the effect of various soil amendments and biostimulants, under fertilized and non-fertilized and irrigated and non-irrigated regimes, on establishment of *Acer rubrum* L. (red maple) and *Crataegus phaenopyrum* (Blume) Hara (Washington hawthorn) liners.

## Materials and Methods

*Plant material.* This experiment was conducted at the Virginia Tech Urban Horticulture Center, Blacksburg, VA (USDA hardiness zone 6a) from May 1995 to October 1996. One hundred, forty-four *Acer rubrum*- L. (red maple), and one hundred, forty-four *Crataegus phaenopyrum* (Blume) Hara (Washington hawthorn) bare-root seedlings were obtained from Lawyer's Nursey, Plains, MO. Seedlings were planted in rows 1m apart in a Groseclose silt loam soil (clayey, mixed, mesic Typic Hapludults), pH 6.2. There was one species per row, and rows alternated between species. Initial height and stem diameter measurements (5cm from ground level) were recorded on 11 May, 1995.

*Statistical design.* The design was a split-split plot design with whole plots (33m x 2m) being irrigated or not irrigated. Split plots were fertilized or not fertilized, and split-split plots were the 6 soil treatments completely randomized within the split plot. Whole plots were replicated two times with 2m spacing between plots. Soil treatments were replicated 3 times within each split plot. Split plots were separated by a 5m buffer zone within each whole plot. Washington hawthorns and red maples were analyzed separately.

*Treatments.* Soil treatments were applied to trees at planting. Planting holes were dug by tractor-powered auger to a depth of 30cm and a diameter of 30cm. Soil treatments were: 1) control; 2) yard-waste compost; 3) Canadian sphagnum peat; 4) granular humate 100g; 5) granular humate 200g; and 6) liquid humate +. The control treatment consisted of the native backfill soil only. The compost (Recycling Center, Virginia Tech, Blacksburg, VA) and peat (Southland Sphagnum peat moss, Greensboro, NC) treatments were applied at a rate of 25:75 amendment:soil (v:v), well mixed before application. Granular humate (Earthgreen Products, Dallas, TX) treatments were surface applied at a rate of 100g/tree, and 200g/tree. Liquid humate+ (Roots™, Roots Inc., New Haven, CT) was applied as a 30 minute root soak (20ml Roots™ in 1L of H<sub>2</sub>O) immediately prior to planting.

All trees were irrigated at planting. Non-irrigated trees were irrigated daily for the first week after transplanting, and irrigation was withheld thereafter. Soil moisture levels were monitored with four gypsum blocks (Irrometer, Riverside, CA) randomly installed 20cm deep within the irrigated plots. Trees were irrigated at a mean soil moisture tension of .05MPa for irrigated trees only using hand irrigation initially, and later a micro-irrigation system. Non-irrigated trees were irrigated only if trees were in danger of desiccation. Additionally, to monitor the soil amendment effect on soil moisture levels in non-irrigated plots, gypsum blocks were installed (15cm deep) within the planting holes of both tree species on three replications of four treatments. Treatments selected were: control, compost, peat, and humate 100g treatments. These amendments were selected because research is limited on how they effect soil water retention around the planting hole in non-irrigated situations. Granular fertilizer (ammonium nitrate 34-0-0, Southern States, Richmond, VA) was applied to split plots at planting at a rate of

4.5g N/m<sup>2</sup>. The entire experiment received fertilizer the following spring on 12 April, 1996 at a rate of 2.3g/m<sup>2</sup>.

*Data collection.* Subsequent stem height and trunk diameter measurements were taken on 9 Sept. 1995, and 21 Aug. 1996 (5cm from ground level). The experiment was terminated on 25 Sept. 1996, at which time tree tops were harvested and dried to a constant mass at 70C. In order to determine treatment effects on root growth, root systems of one whole plot replication (non-irrigated and non-fertilized), resulting in six replications per treatment, was randomly selected. All roots were harvested within a 43cm diameter x 30cm depth volume on each replication. Each root system was separated into five diameter classes (0-1mm, 1-3mm, 3-5mm, 5-10mm, and >10mm), and dried to a constant mass at 70C.

One root system was randomly chosen from each soil treatment for determination of length:dry mass relationship. Root systems were divided into diameter classes as discussed above. Each diameter class was separated into subsamples of varying amounts which were used for determination of the root length:dry mass relationship. Root length of the subsamples was measured using a combination of computer imaging software (Desk-Scan II, Hewlett Packard Co.), and a computer-image analyzing system (Delta-T SCAN, Delta-T Devices LTD. Cambridge, England). Subsamples were then dried to a constant mass at 70<sup>0</sup> C and weighed. A relationship between dry weight and root length for each diameter class was made using a simple-linear regression procedure (SAS vers. 6.08, Cary N.C.) from which regression equations were generated for each root diameter class. These equations were then applied to the remaining replications for root length determinations.

Height and stem diameter data were analyzed by repeated measures multivariate analysis of variance, as well as single degree of freedom contrasts. Top mass was analyzed with the ANOVA procedure, and single degree of freedom contrasts. Root length and soil moisture data were analyzed by one-way ANOVA using Tukey's HSD to separate the mean values (SAS vers. 6.08, SAS Institute, Cary, NC).

## Results

*Top Growth-* Washington hawthorn. Figures 1 and 2 illustrate stem height and diameter growth over two growing seasons, and most of the growth took place during the second season. Contrast analysis over time showed that stem heights of compost treatments were less than for biostimulant treatments (Figure 1, and Table 1a). For all treatments, and granular humate exclusively, stem diameters were higher than for controls. Also, stem diameters of granular humate treatments were higher compared to liquid humate + treatments (Figure 2, and Table 1a). Tests for irrigation, fertilizer, irrigation x fertilizer, and treatment x irrigation x fertilizer effects were not significant for stem height or stem diameter (Table 1a).

Controls and compost-treated trees had lower stem mass than other treatments (Figure 3, and Table 1a). When compared to controls, granular humate treated trees had the higher stem

mass. As seen for stem height and diameter, there were no significant effects of irrigation, fertilizer, irrigation x fertilizer, or irrigation x fertilizer x treatments on stem mass (Table 1a).

*Root Growth* - Washington hawthorn. Most root growth was in the 0-1mm diameter class for all treatments (Table 2). Peat and humate (200g) produced more root length than compost treated trees in the 0-1mm diameter class and for total root length. Humate (200g) produced more root length than compost-treated trees in the 1-3mm diameter class as well. However, no treatment was higher than the control for any diameter class measured, or total root length (Table 2).

*Top Growth*- Red maple. As for Washington hawthorn, the majority of growth for red maple took place during the second growing season. But, contrary to hawthorn, control treatments had the greatest growth overall (Figures 3 & 4, and Table 1b). Height was not affected by any factor tested (Table 1b). However, contrast analysis over time revealed that stem diameters for controls and compost-treated trees were higher compared to other treatments in irrigated but not non-irrigated plots (Figure 6, and Table 4). Similarly, stem mass of compost-treated trees was larger than biostimulant-treated trees, and controls had a larger stem mass than granular humate-treated trees in irrigated but not non-irrigated plots. Liquid humate-treated trees had a lower stem mass than granular humate treatments in non-irrigated plots, but were similar in the irrigated plots (Figure 8, and Table 4).

*Root Length*- Red maple. The majority of root length occurred in the 0-1mm diameter class, with the peat treatment having the highest root length (Table 3). For the 1-3mm class, peat had the highest root length compared to liquid humate+ exclusively. In the other diameter classes there were no differences between treatments. Peat had the highest total root length compared to all other treatments (Table 3).

*Soil moisture levels*. There were no differences between soil treatments for Washington hawthorn in 1995 and 1996 in the non-irrigated plots (Figure 9). However, peat tended to have lower moisture levels than the other treatments. For red maple, moisture levels of control treatments were lower than the other treatments in 1995 (Figure 10). However, in 1996 peat resulted in drier conditions compared to the other treatments. Conversely, granular humate had the highest moisture level of all treatments, but not significantly higher than compost.

## **Discussion**

These results show that soil amendments have some influence on growth of Washington hawthorn. Furthermore, the effects of amendments on stem diameter and stem mass of red maple were influenced by irrigation treatments. However, varying irrigation and fertilizer regimes alone had little impact on post-transplant growth of either tree species.

For both Washington hawthorn and red maple, the majority of top growth took place during the second growing season. This may be a common trend for a tree's recovery after transplanting. Others have found that it may take a number of years for trees to recover from transplant stress (post-transplant shock) and begin to show increases in shoot growth. This has been attributed to plant water stress produced by the reduction of roots during the transplanting process (Watson et. al. 1986; Kjelgren and Cleveland, 1994).

As indicated, Washington hawthorn had a greater growth response to soil amendments than red maple. For Washington hawthorn, top growth was generally lower on controls vs treatments (except height), but for red maple the control treatments resulted in the greatest growth. These results further demonstrate the variability between plant species response to soil amendments. Others have also found differing growth responses to soil amendments depending on the tree species (Corley, 1984).

As previously stated, there was a significant interaction between treatments and irrigation for red maple in regards to stem diameter and mass. Growth was reduced for the control and compost treatments in the non-irrigated treatments. Rainfall amounts for May to October were 60% normal for 1995, and 80% normal for 1996, respectively. The lower than normal rainfall during the first growing season may explain why control trees resulted in less growth in the non-irrigated plots. However, there is not a clear reason why growth was reduced for the compost treatment in the non-irrigated plots. Most likely, an alteration of some factor of soil chemical properties, such as pH (pH of compost was 7.9), but more research is needed to explain this occurrence. Stem diameter and mass were higher for granular humate treatments in the non-irrigated plots compared to the irrigated plots for red maple. One possible explanation for this is that since granular humate was applied as a surface dressing, the humate may have been carried away by surface irrigation run-off, thus reducing its effect on tree growth in the irrigated plots.

Root length was highest for both tree species when the backfill was amended with peat, but only higher than the control for red maple. For both tree species, the majority of the root length occurred in the 0-1mm diameter class, possibly due to the lighter soil texture produced by the addition of peat. However, the treatment that had similar root length to peat was the granular humate treatment (200g). This indicates that factors other than alterations of soil physical properties influenced root growth. Granular humate has shown to increase plant root growth for a number of plant species, including Turkish hazelnut and red maple, as was seen in Chapters 3 and 4. As indicated earlier, humates have been reported to have an auxin-like affect on root growth (O'Donnell, 1973; Tattini et. al., 1991).

The percentage of root growth that occurred outside the planting hole is not known. Do to time constraints, comparisons between original planting hole root density and beyond planting hole density were not determined. Rapid root growth outside of the original planting hole is desirable for good plant establishment. High root lengths, therefore may have been influenced by proliferation of roots within the original backfill. Although a relationship between higher root length within the harvested soil volume and eventual growth beyond the backfill is assumed, long-term monitoring of plant growth would be required for confirmation.

Irrigation treatments did not affect growth of Washington hawthorn or red maple. However, as stated earlier, there was a significant affect of irrigation x treatments on the stem diameter and leaf/stem mass of red maple. Furthermore, the irrigation effect on stem height of red maple was tending towards significance ( $P=0.06$ ). These varied effects between species may be an indication of Washington hawthorn's adaptability to drier soil conditions as compared to red maple.

Overall, the soil moisture levels for Washington hawthorn appeared to be lower than red maple for both growing seasons. One possible explanation for this is a small ground slope affect. The experimental plots were situated in an area with a slight incline, and the planting beds were running across the incline. Washington hawthorn was always planted at the upper side of the incline for each planting bed, thus groundwater movement from the Washington hawthorn to red maples could have caused an increase in red maple soil moisture levels.

The peat treatment tended to have the driest conditions for both species in 1996. Root development was greatest for the peat-treated trees of both species, indicating that increased root absorption area could have resulted in increased water uptake thus depleting the moisture in the soil. Another possibility may be increased evaporation from the soil surface with the peat treatment. There have been reports that peat can sometimes act as a wick and move water to the soil surface for evaporation (Laurie and Ries, 1950).

Fertilizer application at planting did not have a noticeable affect on any growth parameter measured for either species. Similar results have been reported in other studies. For example, Reavis and Whitcomb (1981) noted that the application of fertilizer at planting time of a number of deciduous bare-root species had no effect on tree growth after two growing seasons. Ammonium nitrate, used in this study, is quickly converted to component parts that are easily leached from the root zone. However, the nitrogen rate ( $4.5\text{g N/m}^2$ ) in this study, applied as ammonium nitrate, was two times the level ( $2.25\text{g N/m}^2$ ) typically used for trees. Therefore, nitrogen availability could have been prolonged with this higher rate. There was no indication that fertilizing with nitrogen at the higher rate was harmful to tree growth.

In conclusion, soil amendments, particularly peat and humate, appeared to enhance the growth of Washington hawthorn, but not red maple. Washington hawthorn is considered to be a difficult-to-establish tree (Murakami et. al. 1990; Bates, 1994). Therefore, application of soil amendments at planting of such trees may have some merit. Peat and humate tended to have an increasing affect on root growth. Increased root growth is generally considered advantageous for a tree's survival since it brings more root surface area in contact with the soil for increased water and nutrient uptake.

As was observed in chapters 3 and 4, the one biostimulant with added hormones and vitamins in this study, liquid humate +, did not have positive effect on growth for either tree species. Thus the use of these new products in tree establishment under conditions similar to this study is still highly questionable.

Table 5.1 P- values from repeated measures ANOVA and single degree of freedom contrasts for effects of treatments on height, and stem diameter of Washington hawthorn and red maple, and p-values from ANOVA and contrasts for stem dry mass.

A: Washington Hawthorn	Height (cm)	Stem diameter (mm)	Stem mass <sup>Z</sup> (g)
Source		P > F	
Irrigation	0.14 <sup>Y</sup>	0.23	0.23 <sup>X</sup>
Fertilizer	0.84	0.61	0.79
Fertilizer x irrigation	0.86	0.98	0.94
Treatment	0.04	0.39	0.10
Treatment x irrigation	0.59	0.52	0.60
Treatment x fertilizer	0.55	0.73	0.49
Treatment x fertilizer x irrigation	0.45	0.99	0.46
Contrasts			
Control vs others	0.64	0.04	0.06
Compost vs biostimulants	0.004	0.50	0.01
Peat vs biostimulants	0.15	0.92	0.47
Control vs granular humate	0.20	0.006	0.02
Humate vs Liquid humate+	0.21	0.04	0.21

<sup>Z</sup> Includes leaves.

<sup>Y</sup> P-values from Wilk's Lambda multivariate tests.

<sup>X</sup> P > F

Table 5.1 continued...

B: Red Maple	Height (cm)	Stem diameter (mm)	Stem mass <sup>Z</sup> (g)
Source		P > F	
Irrigation	0.06 <sup>Y</sup>	0.66	0.13 <sup>X</sup>
Fertilizer	0.30	0.66	0.36
Fertilizer x irrigation	0.25	0.78	0.68
Treatment	0.08	0.07	0.03
Treatment x irrigation	0.48	0.02	0.02
Treatment x fertilizer	0.73	0.38	0.64
Treatment x fertilizer x irrigation	0.90	0.92	0.97
Contrasts			
Control vs others	0.08	0.02	0.02
Compost vs biostimulants	0.08	0.25	0.10
Peat vs biostimulants	0.12	0.60	0.45
Control vs granular humate	0.09	0.02	0.05
Humate vs Liquid humate+	0.70	0.88	0.38

<sup>Z</sup> Includes leaves.

<sup>Y</sup> P-values from Wilk's Lambda multivariate tests.

<sup>X</sup> P > F

Table 5.2 Effects of soil treatments on root length (m) of Washington hawthorn for four different diameter classes, and total root length.<sup>Z</sup>

Treatments	Diameter class				Total (m)
	0-1mm	1-3mm	3-5mm	5-10mm	
Control	55.1ab <sup>Y</sup>	3.3ab	1.3	1.5	61.2ab
Compost	21.1b	2.4bc	0.8	1.0	25.3b
Peat	88.2a	4.0ab	1.3	0.5	94.9a
Humate 100g	43.6ab	2.8ab	1.3	1.0	48.8ab
Humate 200g	78.5a	4.87a	1.1	1.6	86.0a
Liquid Humate+	59.0ab	4.4ab	1.2	1.0	65.6ab
Significance	0.0008 <sup>X</sup>	0.02	0.48	0.20	0.0007

<sup>Z</sup> Roots >10mm in diameter excluded from table.

<sup>Y</sup> means in columns followed by the same letter are not significantly different,

Tukey's HSD ( $P \leq 0.05$ ,  $n = 6$ ).

<sup>X</sup>  $P > F$

Table 5.3 Effects of soil treatments on root length (m) of red maple for four different diameter classes, and total root length.<sup>Z</sup>

Treatments	Diameter class				Total (m)
	0-1mm	1-3mm	3-5mm	5-10mm	
Control	46.1b <sup>Y</sup>	3.1ab	1.3	1.5	52.0b
Compost	29.3b	3.7ab	1.4	1.4	35.8b
Peat	165.8a	5.8a	1.5	1.2	174.4a
Humate 100g	57.6b	5.2ab	1.0	1.1	64.9b
Humate 200g	65.4b	3.1ab	1.0	1.3	70.8b
Liquid Humate+	35.2b	2.3b	1.0	1.0	39.3b
Significance	0.0002 <sup>X</sup>	0.01	0.24	0.66	0.0002

<sup>Z</sup> Roots >10mm in diameter excluded from table; n=6.

<sup>Y</sup> means in columns followed by the same letter are not significantly different,

Tukey's HSD( $P \leq 0.05$ , n=6).

<sup>X</sup>  $P > F$

Table 5.4 P- values from single degree of freedom contrasts for the effects of irrigation x soil treatments on stem diameter, analyzed by repeated measures ANOVA, and top mass, analyzed by ANOVA for red maple.

Contrasts	Stem diameter (mm)		Stem mass (g) <sup>Z</sup>	
	Irrigated	Non-irrigated	Irrigated	Non-irrigated
Control vs others	0.04 <sup>Y</sup>	0.40	0.07 <sup>X</sup>	0.14
Compost vs biostimulants	0.008	0.46	0.005	0.25
Peat vs biostimulants	0.93	0.42	0.88	0.28
Control vs humate	0.005	0.89	0.03	0.83
Humate vs liquid humate+	0.39	0.54	0.65	0.04

<sup>Z</sup> Includes leaves.

<sup>Y</sup> P-values from Wilk's Lambda multivariate tests.

<sup>X</sup> P > F

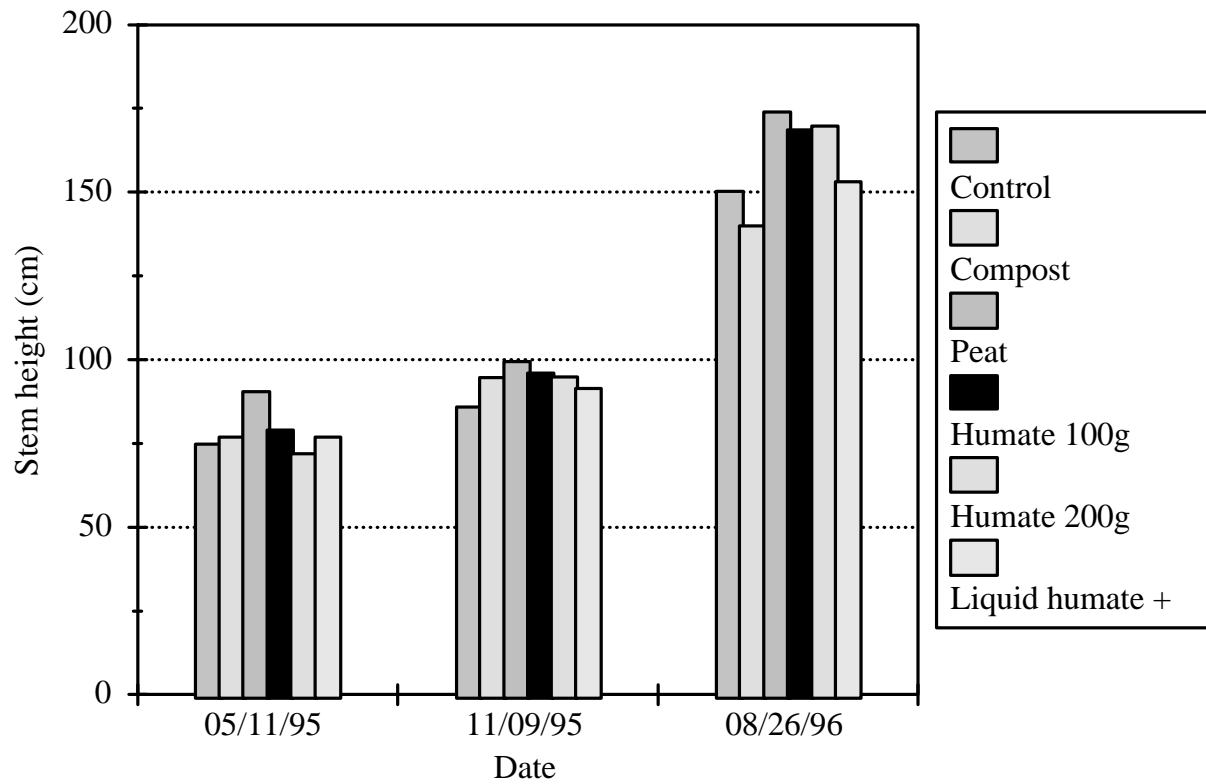


Figure 5.1 Effects of soil treatments on stem height of Washington hawthorn from 11 May, 1995 to 26 Aug. 1996 analyzed with repeated measures multivariate analysis, and single degree of freedom contrasts (Table 1a).

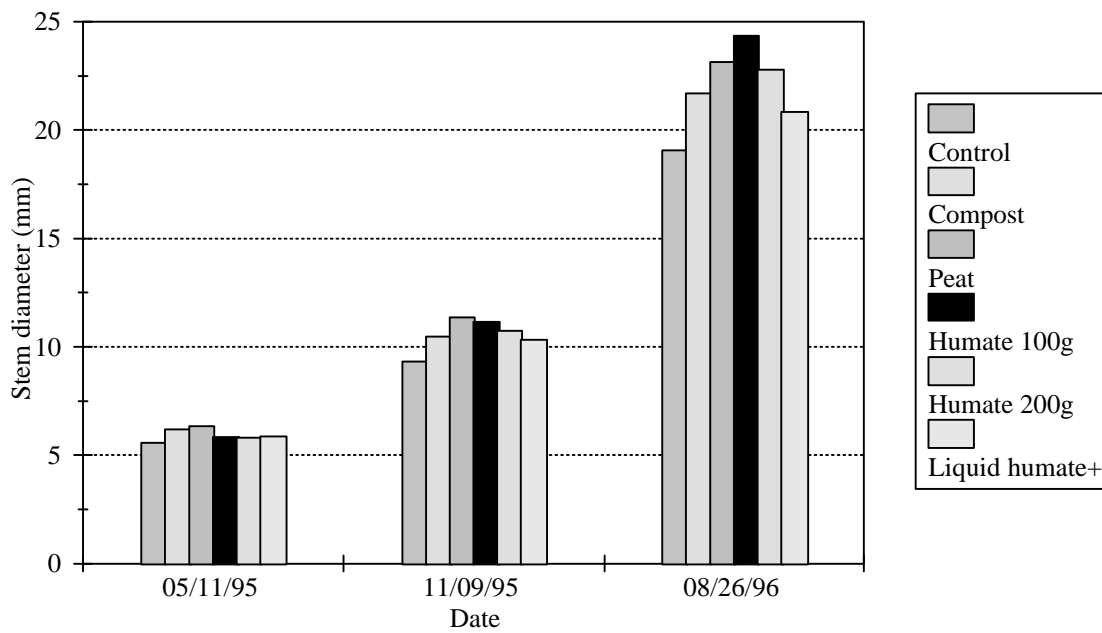


Figure 5.2 Effects of soil treatments on stem diameter growth of Washington hawthorn from 11 May, 1995 to 26 Aug. 1996 analyzed with repeated measures multivariate analysis, and single degree of freedom contrasts (Table 1a).

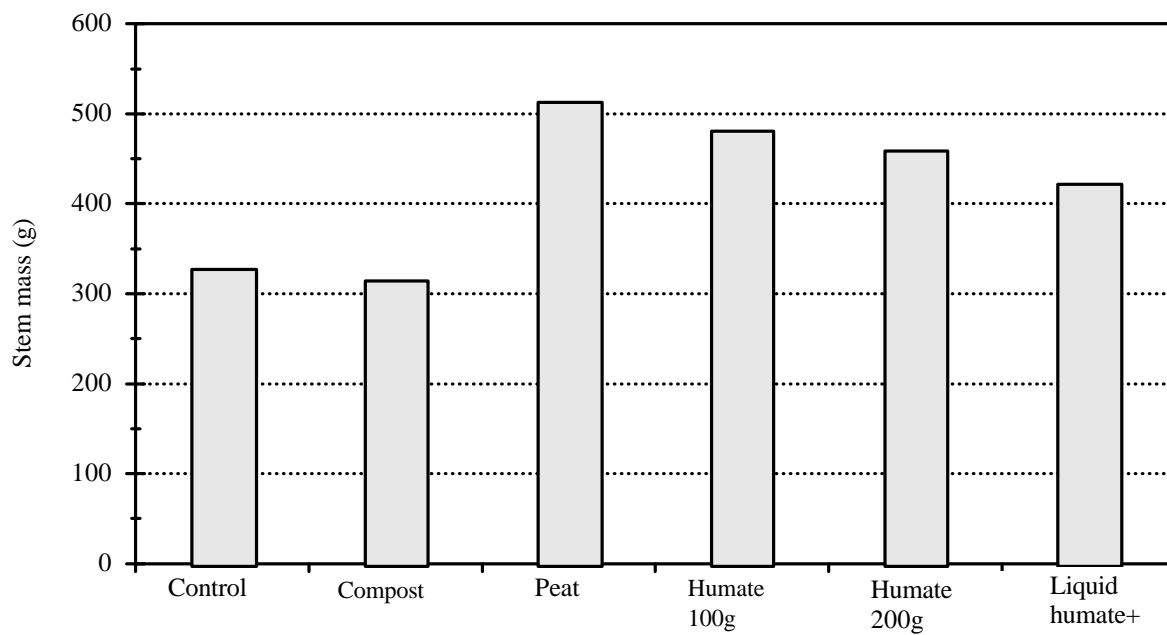


Figure 5.3 Effects of soil treatments on stem mass of Washington hawthorn analyzed by ANOVA, and single degree of freedom contrasts (Table 1a). Standard error values: Control = 50.0; Compost = 45.3; Peat = 55.5; Humate (100g) = 64.5; Humate (200g) = 42.0; and Liquid humate + = 59.0.

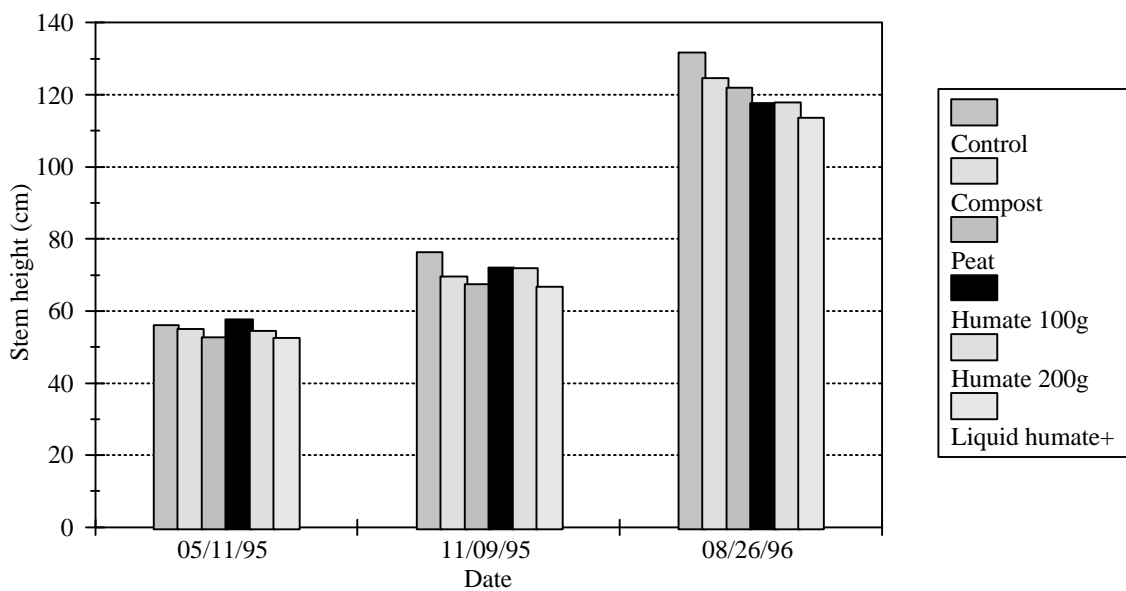


Figure 5.4 Effects of soil treatments on stem height of red maple from 11 May, 1995 to 26 Aug. 1996 analyzed with repeated measures multivariate analysis, and single degree of freedom contrasts (Table 1b).

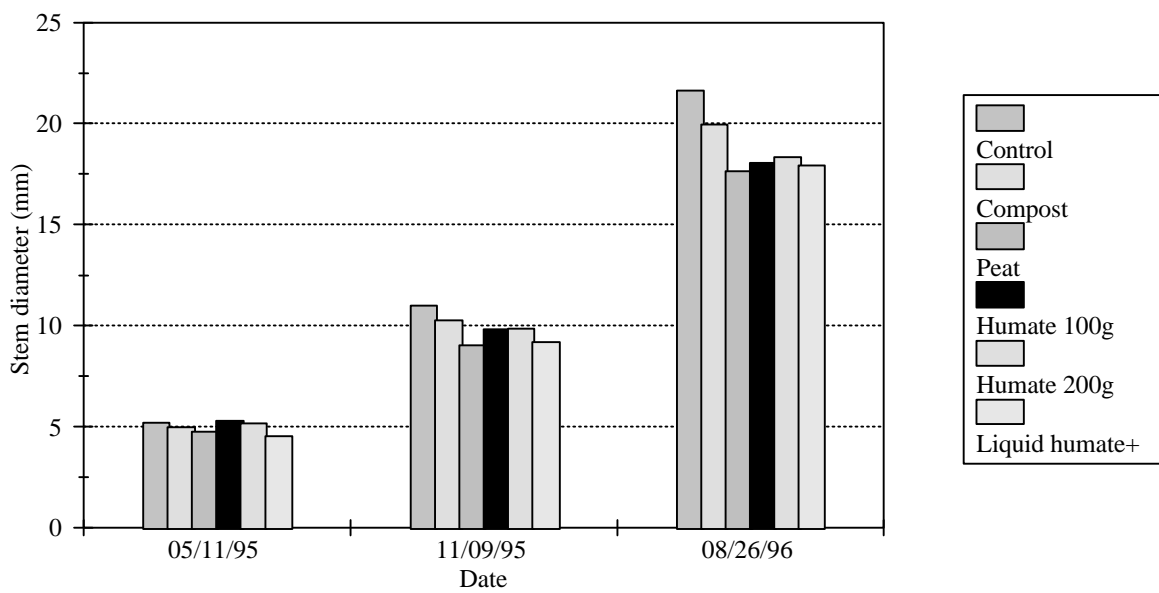


Figure 5.5 Effects of soil treatments on stem diameter of red maple from 11 May, 1995 to 26 Aug. 1996 analyzed with repeated measures multivariate analysis, and single degree of freedom contrasts (Table 1b).

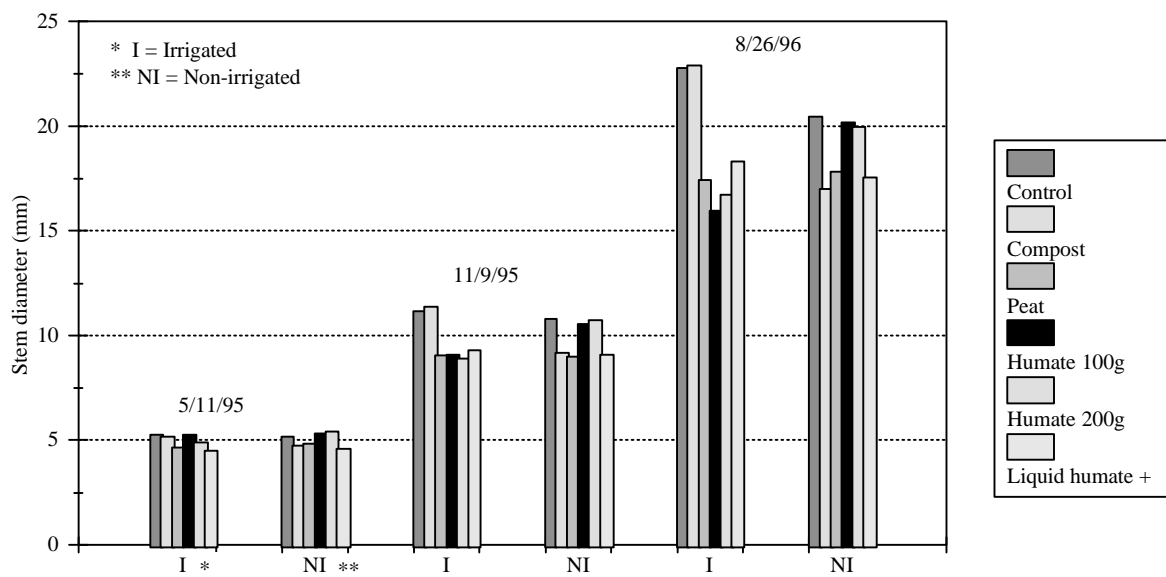


Figure 5.6 Effects of irrigation x soil treatments on stem diameter of red maple from 11 May, 1995 to 26, Aug. 1996, analyzed with repeated measures, and single degree of freedom contrasts (Table 4).

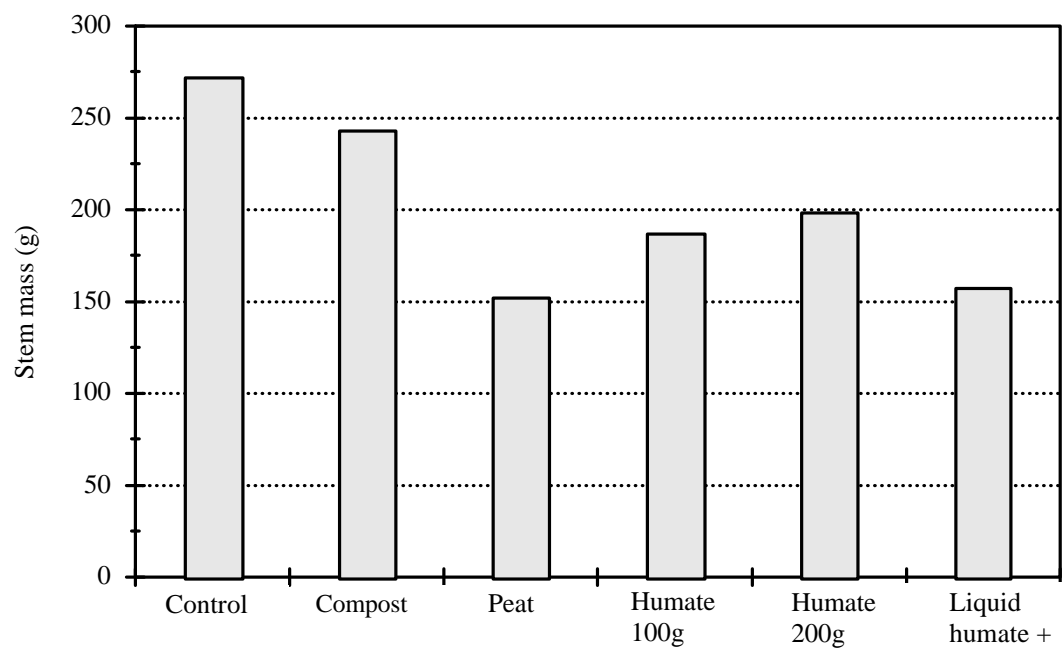


Figure 5.7 Effects of soil treatments on stem mass of red maple analyzed by ANOVA and single degree of freedom contrasts (Table 4). Standard error values: Control = 31.4; Compost = 42.7; Peat = 20.0; Humate (100g) = 33.4; Humate (200g) = 41.2; and Liquid humate + = 28.0.

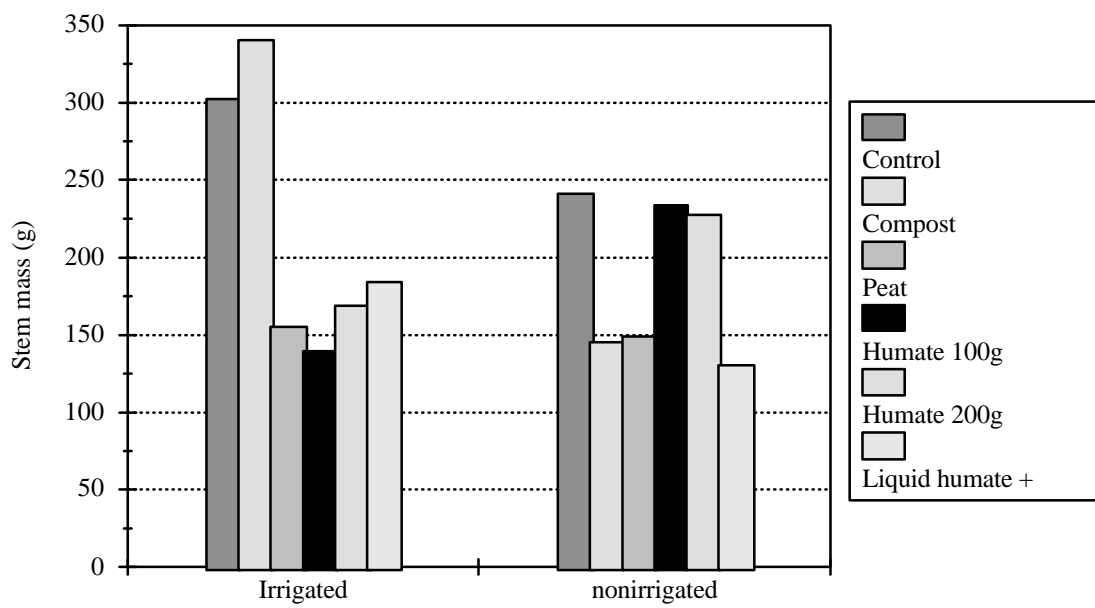


Figure 5.8 Effects of irrigation x soil treatments on stem mass of red maple analyzed by ANOVA, and single degree of freedom contrasts (Table 4).

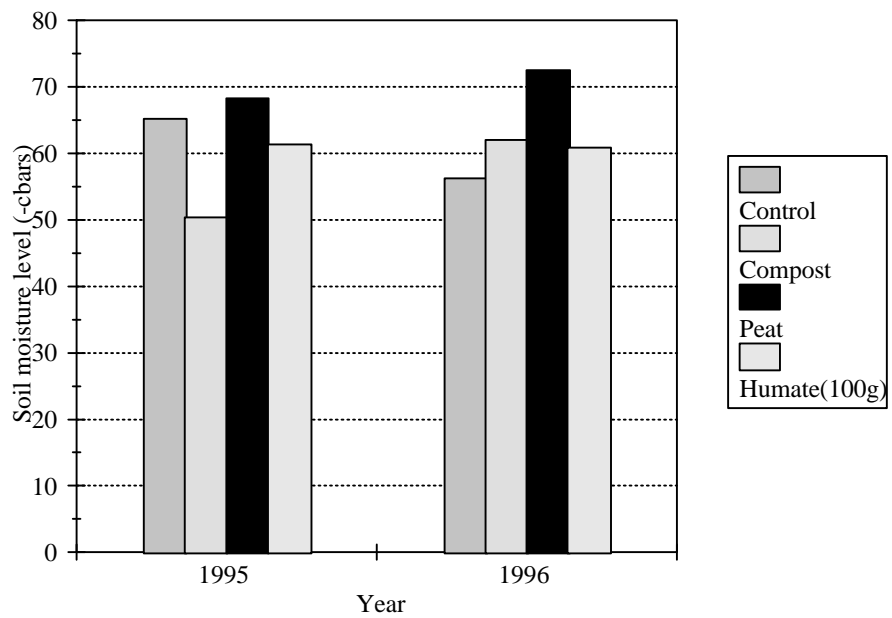


Figure 5.9 Effects of soil treatments on soil moisture level for Washington hawthorn. Treatment means are not significantly different, Tukey's HSD ( $P = 0.05$ ,  $n=3$ ).

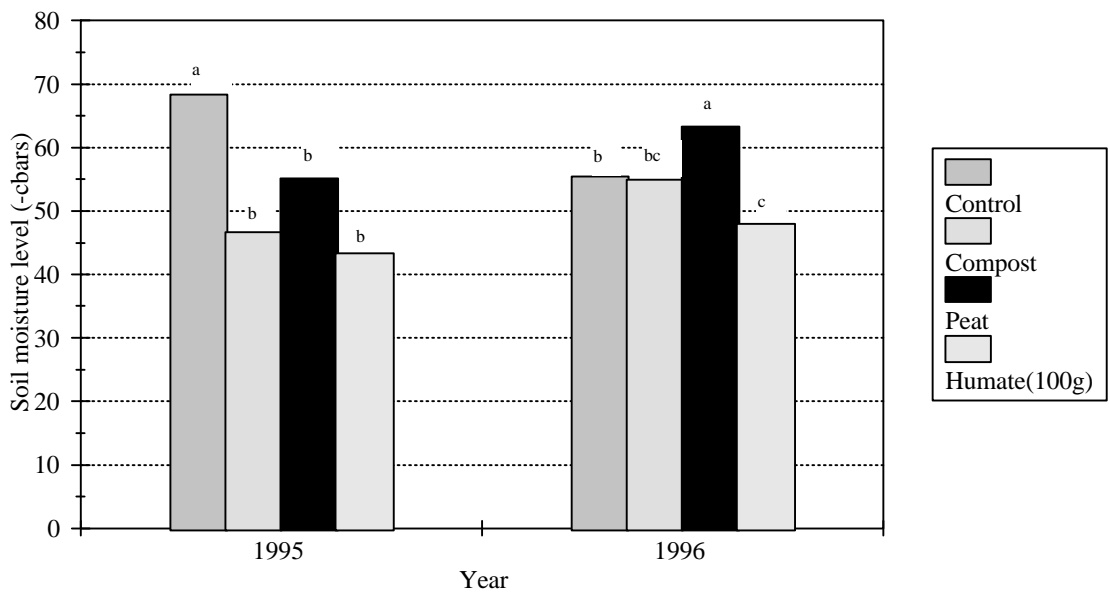


Figure 5.10 Effects of soil treatments on soil moisture level for red maple. Treatment means with the same letter are not significantly different, Tukey's HSD ( $P = 0.05$ ,  $n=3$ ).

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## Chapter Six Conclusion

Results of this thesis indicate that soil amendments and biostimulants offer no consistent advantage to post-transplant tree growth. However, they did result in significantly greater growth than the control treatments for Washington hawthorn (chapter 5), indicating that applying soil amendments to hard-to-establish trees may be worthwhile. Additionally, a trend for increased root growth by granular humate existed, especially at a medium fertilizer rate as seen in the greenhouse experiment with Turkish hazelnut (chapter 3). Biostimulants also had an influence on sap-flow of balled and burlapped red maple soon after transplanting (chapter 4).

There may be several reasons why biostimulants in this study did not consistently increase post-transplant tree growth. Several researchers have exclaimed that the benefits of biostimulant products may be more pronounced in soils with a low organic content (Albregts et. al., 1988; Reynolds et. al., 1995). Throughout the present study, the organic matter of the planting media may have been relatively high, possibly reducing the efficacy of biostimulants. However, other researchers using a planting substrate such as the 50:50 sand:peat mixture used in the rhizotron experiment (chapter 4) have shown humic acids to increase root growth of trees (Tattini et. al., 1991).

Another influential factor may be time of planting. Some researchers have found that certain tree species respond differently to plant hormone applications (a major component of some biostimulants) depending on the season (fall or spring) they were dug, possibly due to shifting endogenous plant hormone levels at the time of harvest (Lumis, 1987). If native hormone levels are high, the effect of exogenous application may be masked. Lastly, shoot growth is sometimes favored over root growth for some species when transplanted in the spring (Watson and Himelick, 1983), and the effect of biostimulants on root growth during this time may be reduced.

In conclusion, soil amendments, such as peat, and granular humate may increase root growth after transplanting in some situations. Furthermore, sapflow was increased by biostimulants soon after transplanting. These two factors are generally believed to be important for successful establishment of trees after transplanting. This research confirms that applications of soil amendments and biostimulants may be beneficial under certain conditions for some species. However, long-term benefits and effectiveness under different conditions (e.g. extreme drought, urban soils, flooded soils) are unknown and should be studied.

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## Vita

Matthew P. Kelting was born on September 13, 1964 in Plainfield, NJ. He and his family moved to Malone, NY in 1969. He graduated from Franklin Academy High School of Malone in 1982, and went on to Onondaga Community College in Syracuse to study music. After several years in Syracuse, Matt returned to upstate NY where he took up a full-time career as a ski instructor at Whiteface Mountain, Lake Placid, NY. Following a number of years of teaching skiing and spending the off-seasons working such jobs as a sound-technician, landscaper, groundskeeper, and maintenance person, Matt returned to college to pursue his bachelors degree in biology at SUNY Plattsburgh, Plattsburgh, NY. Matt got married in 1992, as well as completed his BS degree, and returned to Whiteface for one more season of teaching skiing, and working as a groundskeeper at a Lake Placid resort. At this point, Matt decided that he liked working with plants, especially trees, and wanted to spend his days outside, so he decided to return to school to obtain his MS in horticulture. His brother, Dan, already enrolled at Virginia Tech in College of Forestry graduate program, suggested that Matt come to Virginia Tech because of their excellent horticulture program. In 1994 Matt took his brother's advice, and he and his wife, Ilona, left for Blacksburg, VA where he got a job as a landscaper, and his wife a job as assistant front desk manager at the Donaldson Brown Hotel and Conference Center at Virginia Tech. Matt applied to the graduate school in the fall of 1994, and got accepted into the horticulture graduate program for the spring of 1995. Matt's duties while at VT were working as a GTA, where he assisted in a landscape establishment and maintenance course for a semester, and taught a woody landscape plant identification course for three semesters. He also worked as a caretaker at the Virginia Tech Urban Horticulture Center for the summer months, and as a guest service representative at the Donaldson Brown Hotel. Matt obtained his MS degree in the spring of 1997, and hopes to persue a career in the tree care business, or landscape field.