EFFECTS OF SUBSTRATE, PHENOLOGICAL CUTTING STAGE, AND AUXIN CONCENTRATION ON ROOTING OF *COTINUS OBOVATUS* RAF.

A Senior Honors Thesis

By

GEOFFREY CARLILE DENNY

Submitted to the Office of Honors Programs
& Academic Scholarships
Texas A&M University
In partial fulfillment of the requirements of the

UNIVERSITY UNDERGRADUATE
RESEARCH FELLOWS

April 2000

Group: Cell Biology 1

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April 2000

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ABSTRACT

Effects of Substrate, Phenological Cutting Stage,

and Auxin Concentration

on Rooting of *Cotinus obovatus* Raf. (April 2000)

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A study was conducted to determine the effects of substrate, phenological stage of cuttings, and auxin concentrations on the rooting of Texas smoke tree (*Cotinus obovatus* Raf.). Softwood, semi-hardwood and hardwood cuttings were treated with either a 0 mg/L (ppm), 5000 mg/L (ppm), 10000 mg/L (ppm), or 15000 mg/L (ppm) potassium salts of indolebutyric acid (K-IBA) and placed in either 50% peat : 50% perlite or 100% perlite rooting substrates. Cuttings were placed under an intermittent mist system in a greenhouse for 8 weeks. Softwood cuttings rooted in both substrates, but the 50% peat : 50% perlite substrate produced better quality rooted cuttings. Softwood cuttings peaked at 8,000 to 10,000 mg/L (ppm) K-IBA. Semi-hardwood and hardwood cuttings rooted only in the 100% perlite substrate. In 100% perlite substrate, the optimal concentration for semi-hardwood cuttings was approximately 12000 mg/L (ppm) K-IBA, while hardwood rooting was maximized at 15000 mg/L (ppm) K-IBA or more.

ACKNOWLEDGMENTS

I would like to thank my advisor, Dr. Mike Arnold, who always made time to answer my questions and offer help and guidance with a smile. Thank you to Dr. Wayne Mackay for your advice and assistance. Great thanks to my special helpers, Susan Harvey for all of her help and support and for believing in me, and Lindsey Cappelle for her assistance and friendship. Thanks to my friend Garry McDonald for listening to me and helping me keep my sanity. My thanks to Terry Bacon for allowing me the time to work on my project and for his friendship and understanding. I would also like to acknowledge and thank Dr. Jonathan Egilla for providing me with supplies and greenhouse space. Thanks to my family for their love and support.

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INTRODUCTION

The rapidly increasing human population in Texas and the decreasing water supply necessitate wise use of available water (Welsh et al., 1998). This means conservation, and one of the easier ways for homeowners and businesses to conserve water is by xeriscaping with drought tolerant native plants (Welsh et al., 1998). There is an increasing demand from consumers for low maintenance, drought tolerant plants that are attractive and durable (Wasowski and Wasowski, 1997). There are many native species that meet these criteria and have potential in the landscape trade (Wasowski and Wasowski, 1997). However, many are not readily available at nurseries and garden centers (Goyne and Arnold, 1996). Due to the limited amount of data that exists about the cultural requirements of native species, and with the increasing demand, there is a growing need for research in this area.

Cotinus obovatus Raf. is a large shrub or small tree, growing from 4.6 to 10.7 m (15 to 35 ft.) in height (Vines, 1960). The common names, Texas smoke tree and American smoke tree, refer to the tree's flowers which form purple/pink clouds of filamentous peduncles in mid-spring (Harrar and Harrar, 1946). The leaves are deciduous, simple, obovate, 3.8 to 15.0 cm (1½ to 6 in.) long and 5.0 to 8.9 cm (2 to 3½ in.) wide, and when the leaves emerge in the spring they are colored both pink and lime (Wasowski and Wasowski, 1997). They also show a wide range of fall

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color, from yellow and orange to bright scarlet (Simpson, 1988). The twigs change from green to reddish or purple colors, and eventually gray with age (Vines, 1960). *Cotinus obovatus* grows on well drained, limestone soils and is a good ornamental tree for sites meeting this description (Wasowski and Wasowski, 1997). On these sites, it is often hard to grow other species (Wasowski and Wasowski, 1997). *Cotinus obovatus* has potential as a landscape plant, but vegetative propagation methods have not been fully developed (Mackay, 1999).

Hartman et al. (1997) state that rooting substrates should hold enough moisture to provide sufficient water to the cuttings once mist intervals are reduced. Substrates should be "sufficiently porous so that excess water drains away, permitting adequate penetration of oxygen to the roots" to ensure good rooting (Hartman et al., 1997). Koller and Shadow (1984) report that it is critical to not let the cuttings stay to wet.

Cuttings are treated with auxins to increase rooting percentages, to shorten the rooting period, and to give more uniform rooting (Hartman et al., 1997). In general, the more phenologically mature the tissue, the higher the optimum auxin concentration required to induce rooting (Hartman et al., 1997). At high auxin concentrations, there is the possibility of toxicity, which can lead to poor survival in rooted cuttings (Hartman et al., 1997). Koller and Shadow (1984) recommend Hormodin Number 3 (8000 mg/L (ppm) IBA) for use on softwood cuttings.

The limited information available on vegetative propagation of *C. obovatus* suggests the need for investigation of the effects of different substrates, physiological stage of the cutting, and auxin concentration. Therefore, the objective of this study was to develop a vegetative propagation system for *C. obovatus*. The null hypothesis is that the type of cutting, auxin concentration, and media type will have no effect on the rooting of *C. obovatus*.

MATERIALS AND METHODS

Cuttings at three phenological states (softwood, semi-hardwood, and hardwood) were collected on 20 March 1999 at the Texas Agricultural Research and Extension Center at Dallas. The softwood was defined as new, unlignified growth, semi-hardwood as tissue that is beginning to mature and undergo lignification, and hardwood as lignified previous season's growth. The cuttings were transported in sealed plastic bags by truck and stored in a 4°C (39 °F) cooler until treated. On 27 March 1999, the cuttings were trimmed to 12.5 to 15.0 cm (5 to 6 in.) with an approximate 45° angle at the base of the cutting immediately beneath a node. The lower four nodes were stripped of leaves. The cuttings were submerged for approximately 7 seconds in a 50 mg/L (ppm) bromine solution to help prevent algal, fungal, and bacterial growth. Afterward, the bottom 5.0 cm (2 in.) of the cuttings were treated with different concentrations of K-IBA: 0 mg/L (ppm), 5000 mg/L (ppm), 10,000 mg/L (ppm), or 15,000 mg/L (ppm). A 0 mg/L (ppm) K-IBA treatment with the bromine treatment omitted was included. Fifteen of the cuttings at each concentration level were placed into 50 cell (5.0 cm x 6.0 cm (2 x 2 in.)) plug sheets (Landmark Plastic Corporation, Akron, Ohio) filled to level with the 1 part sphagnum peat (Sun Gro Horticulture, Pine Bluff, Arkansas) : 1 part coarse perlite (Strong-Lite® Products Corp., Pine Bluff, Arkansas) (by volume) rooting mix with 1 cutting per cell and five cuttings per block. The cuttings were inserted by pushing them directly into the substrate to a depth of 6.0 cm (2 in.). Fifteen cuttings were identically placed into 50 cell plug sheets filled with 100% coarse perlite. Sheets containing a given substrate were randomly intermixed within a block.

These two substrates were chosen because the 1 part sphagnum peat : 1 part coarse perlite (by volume) is a conventional rooting substrate with high water holding capacity which is based on the Cornell Peat-Lite mixes (Hartman et al., 1997). Type 2 is 100% coarse perlite, a substrate that provides good drainage and aeration (Hartman et al., 1997). Goyne reports rooting in low percentages using a 3 parts fine screened pine bark : 1 part builders sand (by volume) (Goyne, 1998).

Cuttings were placed immediately in a greenhouse under an intermittent mist system (6 sec. each 8 min., dawn to dusk) using reverse osmosis water for 8 weeks. The treatments were monitored weekly to determine if rooting had occurred. The cuttings were harvested on 22 May 1999 because there was significant rooting in most treatments and non-rooting treatments were beginning to deteriorate. At harvest, the number of cuttings rooted out of 5 and the mean number of roots per cutting were determined. The mean of the shortest and the longest roots per cutting was determined and a mean for each replication generated. Fresh mass of the roots per replication was measured. Root samples were dried in an oven (70°C (158°F) for 60 days) and the dry root masses were measured.

The plug sheets were placed in the greenhouse in a randomized complete block (2 substrate x 3 cutting types x 5 K-IBA treatments x 3 blocks with 5 cutting replications per block). In the initial analysis the 0 mg/L (ppm) ± bromine treatments were compared to determine the effects of bromine utilization. After pooling the 0 mg/L (ppm) treatments, analysis of variance were conducted for the remaining factors (SAS Institute, 1988). Means, standard errors and step-wise polynomial regression equations were generated for significant effects. Only interactions or main effects not involved in significant ($P \leq 0.05$) higher order interactions are presented.

RESULTS AND DISCUSSION

Controls did not differ significantly (paired t-test, $P \leq 0.05$), therefore, 0 mg/L (ppm) ± bromine were pooled to evaluate the 0 mg/L K-IBA treatment effects in subsequent analysis. Third order interactions among substrate type, cutting type and auxin concentration were significant ($P \leq 0.05$) for all rooting measures. Hence, lower order interaction and main effects are not presented. Data indicated similar results for both dry and fresh mass, therefore only dry mass data are presented.

Softwood cuttings were the only type of cutting that had significant rooting in the 50% peat : 50% perlite (by volume) substrate, where as semi-hardwood and hardwood did not (Fig. 1.). This may have been due to the higher water holding capacity of the substrate compared to 100% perlite. The younger, suppler, tissue may have benefited from the extra moisture, but may have been excessive for the more mature tissues. This would be in agreement with Koller and Shadow (1984), who recommend not allowing cuttings to get too wet during propagation. Also, Hartman et al. (1997) suggest that excessive water in the rooting media is detrimental to root formation.

Softwood cuttings in both substrates had the highest number of rooted cuttings per five at approximately an 8,000 to 10,000 mg/L (ppm) K-IBA concentration (Fig. 1, Table 1). This is in agreement with Koller and Shadow's (1984) recommendation of 8,000 ppm IBA with softwood cuttings. The softwood, 50% peat : 50% perlite treatment had significantly higher numbers of roots per cutting (Fig. 2, Table 1), longer roots (Fig. 3, Table 1), and a greater root dry masses (Fig. 4, Table 1) than the softwood, 100% perlite treatment, when auxin was applied. This may be due in part to the more damaging nature of the 100% perlite substrate to the softer tissue during the insertion process and increased moisture content in the 50% peat : 50% perlite substrate. The softwood, 50% peat : 50% perlite treatment had significantly longer roots than the other treatments (Fig. 3) and a larger dry mass (Fig. 4). This may again be the result of the increased root available moisture in the substrate.

Semi-hardwood, 100% perlite treatment had the maximum number of rooted cuttings per five (Fig. 1.), more roots per cutting (Fig. 2.), and greater root dry masses (Fig. 4) at concentrations of approximately 12,000 to 15,000 mg/L (ppm) K-IBA. The same substrate and cutting type had peak root length at a lower concentration, approximately 8,000 to 10,000 mg/L (ppm) K-IBA (Fig. 3).

Peak rooting of the hardwood, 100% perlite in terms of number of rooted cuttings per five (Fig. 1.), number of roots per cutting (Fig. 2.), root length (Fig. 3), and dry mass (Fig. 4), occurred at 15,000 mg/L (ppm), however an optimal concentration was not determined as concentrations in excess of 15,000 ppm were not tested.

These results for both the semi-hardwood and hardwood treatments follow a conventional trend; more mature tissue normally requires a higher auxin concentration to root significantly.

CONCLUSIONS

Propagation of softwood can be achieved in either substrate with a concentration of 8,000 mg/L (ppm) K-IBA, however, the 50% peat : 50% perlite substrate produces higher quality rooted cuttings. When using 100% perlite, holes should be made in the substrate prior to inserting the cutting to prevent damaging the tissue and possibly improve rooted cutting quality.

When propagating semi-hardwood or hardwood cuttings, the better-drained substrate is critical. To obtain peak rooting, a K-IBA solution of approximately 12,000 mg/L (ppm) should be used for semi-hardwood and at least a 15,000 mg/L (ppm) K-IBA solution for hardwood cuttings in 100% perlite substrate.

Survival studies were not performed on any of the rooted material due to the destructive nature of the rooting measurements, therefore any possible detrimental effects of the higher concentrations of K-IBA could not be assessed. Additional examination and experimentation in this area would be beneficial. A need exists for further studies to determine the effects of possible mechanical damage during cutting insertion, as well as the utilization of other forms of rooting containers. Stock plants were growing in landscape settings, with not additional fertility applied. The physiological conditions of stock plants have been shown to impact rooted cutting performance (Rowe et al., 1996), hence studies with better tended stock plants might yield higher quality rooted cuttings.

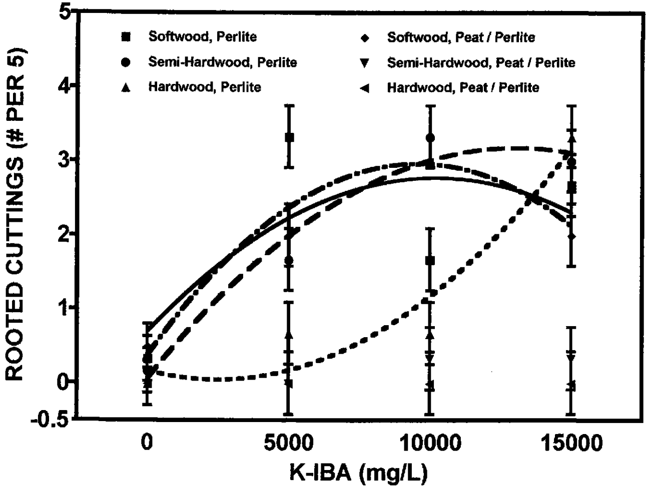

Figure 1. Means (± standard errors) and second order polynomial regression equations for the number of cuttings rooted out of five for softwood (■, —, $P \leq 0.03$, $R^2 = 0.46$), semi-hardwood (•, -----, $P \leq 0.01$, $R^2 = 0.79$), or hardwood (π, ·····, $P \leq 0.01$, $R^2 = 0.85$) cuttings in 100 % perlite or softwood (◆, -·-·-·-, $P \leq 0.01$, $R^2 = 0.56$), semi-hardwood (θ, ns regression), or hardwood (τ, ns regression) cuttings in 50 % perlite : 50 % peat moss substrate, n = 3. Polynomial regression equations are presented in Table 1.

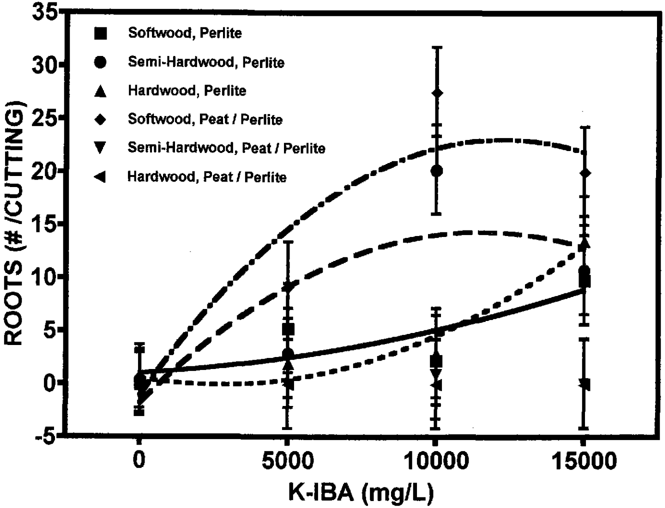

Figure 2. Means (± standard errors) and second order polynomial regression equations for the number of roots per cutting for softwood (■, —, $P \leq 0.08$, $R^2 = 0.36$), semi-hardwood (•, -----, $P \leq 0.09$, $R^2 = 0.33$), or hardwood (π, ·····, $P \leq 0.01$, $R^2 = 0.64$) cuttings in 100 % perlite or softwood (◆, -·-·-·, $P \leq 0.08$, $R^2 = 0.37$), semi-hardwood (θ, ns regression), or hardwood (τ, ns regression) cuttings in 50 % perlite : 50 % peat moss substrate, n = 3. Polynomial regression equations are presented in Table 1.

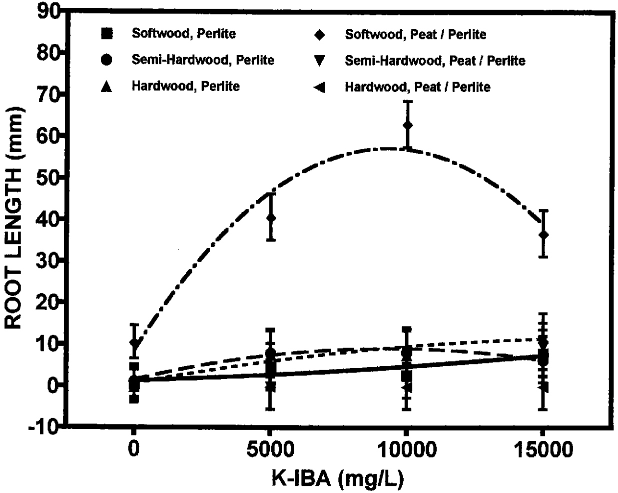

Figure 3. Means (± standard errors) and second order polynomial regression equations for the root length for softwood (■, —, $P \le 0.05$, $R^2 = 0.43$), semi-hardwood (•, -----, $P \le 0.01$, $R^2 = 0.62$), or hardwood (π, ·····, $P \le 0.01$, $R^2 = 0.51$) cuttings in 100 % perlite or softwood (◆, -·-·-·-, $P \le 0.02$, $R^2 = 0.5$), semi-hardwood (θ, ns regression), or hardwood (τ, ns regression) cuttings in 50 % perlite : 50 % peat moss substrate, n = 3. Polynomial regression equations are presented in Table 1.

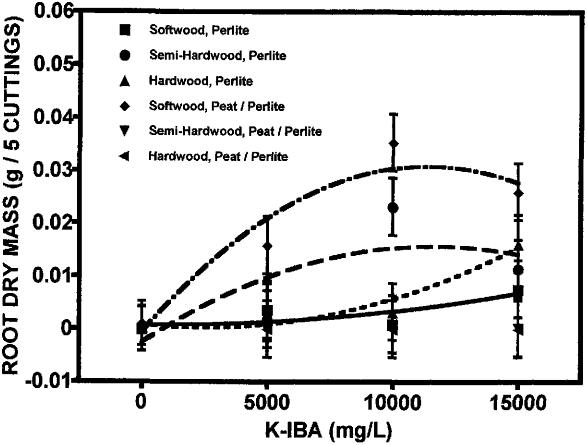

Figure 4. Means (± standard errors) and second order polynomial regression equations for the total root dry mass for softwood (■, —, $P \le 0.13$, $R^2 = 0.31$), semi-hardwood (•, -----, $P \le 0.13$, $R^2 = 0.29$), or hardwood (π, ·····, $P \le 0.01$, $R^2 = 0.58$) cuttings in 100 % perlite or softwood (◆, -·-·-·-, $P \le 0.07$, $R^2 = 0.38$), semi-hardwood (θ, ns regression), or hardwood (τ, ns regression) cuttings in 50 % perlite : 50 % peat moss substrate, n = 3. Polynomial regression equations are presented in Table 1.

Table 1. Second order polynomial regression equations for Figures 1 through 4.

Fig. value	Rooting measure	Substrate	Cutting type	Intercept	(mg/L K-IBA)^1	(mg/L K-IBA)^2	R^2	P
1	Number of cuttings rooted out of five	100% perlite	Softwood	$2.35x10^{-1\,z}$ $5.79x10^{-1\,y}$	$5.10x10^{-4}$ $2.15x10^{-4}$	$-2.49x10^{-8}$ $1.00x10^{-8}$	0.46	0.034
1	Number of cuttings rooted out of five	100% perlite	Semi-hard	$1.11x10^{-1}$ $3.05x10^{-1}$	$4.67x10^{-4}$ $1.20x10^{-4}$	$-1.78x10^{-8}$ $1.00x10^{-8}$	0.79	0.001
1	Number of cuttings rooted out of five	100% perlite	Hardwood	$8.55x10^{-2}$ $2.24x10^{-1}$	$-8.21x10^{-5}$ $8.81x10^{-5}$	$1.91x10^{-8}$ $1.00x10^{-8}$	0.85	0.001
1	Number of cuttings rooted out of five	50% peat / 50% perlite	Softwood	$3.27 x 10^{-1}$ $4.74 x 10^{-1}$	$5.52 x 10^{-4}$ $1.76 x 10^{-4}$	$-2.88 x 10^{-8}$ $1.00 x 10^{-8}$	0.56	0.011
1	Number of cuttings rooted out of five	50% peat / 50% perlite	Semi-hard	ns ns	ns ns	ns ns	ns	ns
1	Number of cuttings rooted out of five	50% peat / 50% perlite	Hardwood	ns ns	ns ns	ns ns	ns	ns
2	Number of roots per cutting	100% perlite	Softwood	$5.77x10^{-1}$ 2.05	$2.66x10^{-4}$ $7.62x10^{-4}$	$1.92x10^{-8}$ $5.00x10^{-8}$	0.37	0.079
2	Number of roots per cutting	100% perlite	Semi-hard	$-7.64x10^{-1}$ 4.18	$2.57x10^{-3}$ $1.65x10^{-3}$	$-1.10x10^{-7}$ $1.10x10^{-7}$	0.33	0.094
2	Number of roots per cutting	100% perlite	Hardwood	$2.75x10^{-1}$ 1.66	$-4.02x10^{-4}$ $6.54x10^{-4}$	$8.32x10^{-8}$ $4.00x10^{-8}$	0.64	0.002
2	Number of roots per cutting	50% peat / 50% perlite	Softwood	$-2.61x10^{-1}$ 6.38	$3.75x10^{-3}$ $2.36x10^{-3}$	$-1.52x10^{-7}$ $1.60x10^{-7}$	0.37	0.077
2	Number of roots per cutting	50% peat / 50% perlite	Semi-hard	ns ns	ns ns	ns ns	ns	ns

2	Number of roots per cutting	50% peat / 50% perlite	Hardwood	ns ns	ns ns	ns ns	ns	ns
3	Root length	100% perlite	Softwood	5.77×10^{-1} 2.05	2.66×10^{-4} 7.62×10^{-4}	1.92×10^{-8} 5.00×10^{-8}	0.37	0.079
3	Root length	100% perlite	Semi-hard	1.44 1.11	1.64×10^{-3} 4.37×10^{-4}	-8.76×10^{-8} 3.00×10^{-8}	0.62	0.003
3	Root length	100% perlite	Hardwood	3.06×10^{-1} 2.03	1.32×10^{-3} 8.00×10^{-4}	-3.91×10^{-8} 5.00×10^{-8}	0.51	0.014
3	Root length	50% peat / 50% perlite	Softwood	10.07 9.34	1.02×10^{-2} 3.47×10^{-3}	-5.49×10^{-7} 2.30×10^{-7}	0.50	0.021
3	Root length	50% peat / 50% perlite	Semi-hard	ns ns	ns ns	ns ns	ns	ns
3	Root length	50% peat / 50% perlite	Hardwood	ns ns	ns ns	ns ns	ns	ns
4	Root Dry Mass	100% perlite	Softwood	4.89×10^{-4} 1.78×10^{-3}	2.41×10^{-8} 6.60×10^{-7}	2.64×10^{-11} 0.00	0.31	0.125
4	Root Dry Mass	100% perlite	Semi-hard	-1.29×10^{-3} 5.14×10^{-3}	2.86×10^{-6} 2.02×10^{-6}	-1.22×10^{-10} 0.00	0.28	0.131
4	Root Dry Mass	100% perlite	Hardwood	4.05×10^{-4} 2.21×10^{-3}	-3.89×10^{-7} 8.70×10^{-7}	9.24×10^{-11} 0.00	0.58	0.005
4	Root Dry Mass	50% peat / 50% perlite	Softwood	9.92×10^{-5} 8.17×10^{-3}	5.37×10^{-6} 3.03×10^{-6}	-2.35×10^{-10} 0.00	0.38	0.071
4	Root Dry Mass	50% peat / 50% perlite	Semi-hard	ns ns	ns ns	ns ns	ns	ns

4	Root Dry Mass	50% peat / 50% perlite	Hardwood	ns ns	ns ns	ns ns	ns	ns

zCoefficient.
yStandard error of coefficient.

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CAREER OBJECTIVE:

To obtain a position with a high degree of responsibility and independence in the field of plant introduction or public gardening.

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G.P.A. - Overall 3.939 Major 4.000

EXPERIENCE:

- **Intern** El Paso Zoo Jan. 2000-Present
  Developed data base for plant accessioning. Exhibit and grounds maintenance. Assisted with planting designs for new and existing exhibits. Plant labeling and mapping. Greenhouse production and maintenance.
- **Field Operations Foreman** Texas A&M Rose and Stone Fruit Breeding Program Mar. 1998-Sep. 1999
  Maintained trials for evaluation roses and stone fruit in the field and greenhouse. Consulted on cultural methods and supervised and trained other workers. Assisted with selection and evaluation of fruit samples and roses.
- **Intern** Texas Agricultural Experiment Station at El Paso May–Aug. 1997
  Constructed and programmed weather station. Maintained plant trials and experimental plant materials. Repaired and installed irrigation systems. Assisted in experimental organization and collected data.
- **Nurseryman** Pearson's Tree Place March 1994- May 1996
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ACTIVITIES:

- **Floriculture-Horticulture Society**
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  1st Place - '98 National Fruit and Nut Judging Contest
- **Attended '99 and '00 Southern Region Meetings of ASHS**
  1st Place - '00 J. B. Edmond Undergraduate Paper Competition

HONORS:

- Phi Eta Sigma Honor Society
- Alpha Zeta Honor Fraternity
- Tommie Vaughn Endowed Scholarship
- Texas Association of Nurserymen Scholarship
- Outstanding Freshman Scholarship
- Mountain Plains Garden Club Scholarship
- Bartlett Tree Foundation Scholarship
- William C. Welch Landscape Scholarship
- The Park People/ Denton A. Cooley Foundation Scholarship