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Effect of Cutting Time and Auxin Application Method on Propagation of *Magnolia grandiflora* 'Southern Charm'

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Summary

Use of foliar auxin applications are increasing in the nursery and greenhouse industry. However, previous research has shown that insufficient auxin is being absorbed or translocated to the site of action. Research was conducted to determine whether cutting time, addition of surfactant to the auxin solution, or auxin application method had an impact on the propagation of *Magnolia grandiflora* 'Southern Charm'. Terminal cuttings of 'Southern Charm' magnolia (*M. grandiflora* 'Southern Charm') were taken at two times of the year: spring and fall and sprayed to the drip point using a solution of Hortus IBA Water Soluble Salts™ at concentrations of 0, 500, 1,000 or 1,500 ppm or dipped for 3-sec in a solution of 2,500 ppm IBA. For many of our tested parameters, fall cuttings were better than spring cuttings. Fall cuttings treated with 1,500 ppm foliar IBA solution plus 0.85 ppm Regulaid® had greater root numbers than spring cuttings treated with less than 1,500 ppm IBA with or without Regulaid®, or fall cuttings treated with a 2,500 ppm basal quick dip, a foliar application of 1,000 ppm or less without Regulaid®, or fall cuttings receiving a foliar application of 0.85 ppm Regulaid®.

INTRODUCTION

Research into foliar auxin applications methods over the past decade indicated that one-time applications are the industry standard (Blythe et al., 2007; Kroin, 2014). When applied post-sticking, much lower concentrations (50 to 100 ppm) of rooting hormones are required compared to other conventional application methods (Dole and Gibson, 2006). Overhead applications of water soluble IBA are increasing in the nursery industry. Bailey Nurseries Inc. in Minnesota and Oregon has been conducting repetitive on-farm trialing for the fifteen years. Their results indicated that many of the taxa commonly propagated respond similarly to foliar-applied auxin compared to a traditional basal quick-dip. At Bailey Nurseries, propagation trays and beds are treated with a single application of a water-soluble IBA solution ranging from 250 to 2,000 ppm (Drahn, 2007).

Decker's Nursery in Ohio uses a battery-powered backpack sprayer to treat their cuttings since it atomized the auxin solution similarly to the mist from the mist system and applied a very small droplet with excellent coverage over both the top and bottom of the cutting (Decker, 2016). Since propagation areas vary in size, overhead applications are applied via a backpack sprayer for small houses and reel-and-hose sprayers for larger production areas.

When being applied overhead, Kroin (2014) of Hortus USA recommends to "spray the solution evenly over the cuttings until drops fall onto the media". To do this, Bailey Nurseries aims to deliver 1 L of solution per 60 ft^2. Currently, both Decker's Nursery and Bailey Nurseries generally treat their cuttings within 24-h of being stuck, either at the end of each day or the following morning, but application occurring during the day in conjunction with frequent mist intervals has not reduced efficacy (Drahn, 2007; Decker, 2016). Cuttings are treated in the early morning or late afternoon due to both lower light levels and reduced misting requirements. For both nurseries, the switch to overhead auxin application led to a decrease in plant material handling and the time cuttings spend in cold storage and the preparation room, where problems associated with lengthened exposure to low temperatures, high humidity, and/or handling can occur (Drahn, 2007). In 2003, 99.6% of cuttings at Bailey Nurseries were quick dipped and 0.4% were treated with foliar applications. By 2007, the percentages had reversed, with 95% of all propagated material being treated with overhead applications and 5.2% of material being quick-dipped (Drahn, 2007). Currently, overhead applications of water-soluble IBA are used to treat the following genera at Bailey Nurseries Minnesota operation: *Acer, Berberis, Cornus, Diervilla, Euonymus, Forsythia, Hydrangea, Juniperus, Lonicera, Philadelphus, Physocarpus, Rhus, Rosa, Spiraea, Symphoricarpos, Syringa, Thuja, Viburnum, and Weigela*. (Drahn, 2007).

Surfactants are common in agricultural production as penetration of the leaf cuticle is required for efficacy of foliar-applied compounds (Robertson and Kirkwood, 1969). Effectiveness of foliar-applied compounds depends on its ability to penetrate through the cuticle and translocate to the site of action (White et al., 2002). Surfactants enhance penetration of these chemicals by increasing the wetting capacity up to the critical micelle concentration (CMC), defined as the concentration above which any added surfactant molecules appear with high probability as micellar aggregates (Ruckenstein and Nagarajan, 1975; Lownds et al., 1987). Research was conducted by Lownds et al. (1987) to determine the effects surfactants would have on foliar penetration of NAA and NAA-induced ethylene production by cowpea [*Vigna unguiculata (*L.) Walp. subsp. *unguiculata* cv. Dixielee]. This research indicated that foliar penetration of NAA was increased when co-applied with a surfactant (Pace®, Regulaid®, or Tween 20®) and all three induced similar qualitative changes in surface tension, contact angle, and droplet: leaf interaction. All three surfactants increased the droplet: leaf ratio. However, Regulaid® was the only surfactant tested that showed a correlation between NAA penetration and interface area (Lownds et al., 1987).

When choosing a surfactant for plant production, several factors should be considered: (1) it should be non-toxic to both the plant and the environment; (2) it should be a small molecule that is water soluble; (3) it should be non-ionic; (4) and relatively effective at decreasing surface concentration at a relatively low concentration (Colwell and Rixon, 1961). While anionic and cationic surfactants are labeled and frequently used in plant production (Dobozy and Bartha, 1976), using non-ionic surfactants are preferable since they do not affect water hardness, nutrient balance, or enzymatic activity and are compatible with most herbicides due to lack of activity with the foliar-applied chemical (Bayer and Foy, 1982).

The objective of this research was to evaluate whether addition of surfactants to foliar auxin solutions increased root growth and uniformity compared to the industry-standard basal quick dip for 'Southern Charm' magnolia (*Magnolia grandiflora* 'Southern Charm').

MATERIALS AND METHODS

An experiment was performed to evaluate the effect of four foliar auxin concentrations [0, 500, 1,000, and 1,500 ppm IBA indole-3-butyric acid (IBA) (Hortus IBA Water Soluble Salts™; Phytotronics Inc., Earth City, MO)] each at two concentrations (0 and 85 ppm Regulaid®) on rooting of 'Southern Charm' magnolia. Additionally, a basal quick dip of 2,500 ppm IBA was used as an industry-based control. Five-node terminal cuttings of *Magnolia grandiflora* 'Southern Charm' [12.7cm (5-in)] were harvested from established landscape plants and stuck to a depth of 1.3 cm (0.5-in) on two dates: 14 April 2021 and 15 November 2021. During cutting preparation, a 2.5 cm (1-in) wound was applied to opposite sides of the basal end of the cutting. Propagation medium was 100% pine bark placed into 8.9 cm (3.5-in) square production pots (T.O. Plastics, Inc., Clearwater, MN). Cuttings receiving foliar applications of auxin were sprayed once to runoff with a 3.75-L battery operated sprayer (One World Technologies, Inc., Anderson, SC). Pine bark for this experiment was sourced from Eakes' Nursery Supply (Seminary, MS) and delivered as a mix of 50% aged and 50% fresh bark passed through a 3/8" (0.95 cm) screen. After treatment, cuttings were placed under intermittent mist applied for 4 sec/4 min during daylight hours and adjusted as needed for the duration. Experimental design was a completely randomized design. Treatment structure was a complete factorial (5 auxin rates × 2 surfactant concentrations × season). Data collected after 120 days included rooting percentage, shoot height, total root number, average root length (three longest roots), and root quality (1-5, with 1=callused cuttings without roots and 5= ≥ 10 roots). Additionally, net photosynthetic rate (A) and stomatal conductance (g_{sw}) values were sampled between the hours of 7:30 A.M. and 11:30 A.M. using the LiCOR 6000 Portable Photosynthesis System (LI-COR Biosciences; Lincoln, NE).

Data were analyzed using linear models and generalized linear models with the GLIMMIX procedure of SAS (ver. 9.4; SAS Institute Inc., Cary, N.C.) by first testing for an interaction between treatment factors (auxin rate, surfactant concentration, and season). When the three-way interaction was significant, auxin rates were compared within surfactant concentration by season. When the three-way interaction term was not significant ($p > 0.10$), main effects means for levels within each treatment factor were compared. Mean separation was performed using the Holm-Simulated Method for multiple comparisons to maintain an overall significance level of $\alpha = 0.05$.

RESULTS

Rooting percentage of Teddy Bear® Southern magnolia (*Magnolia grandiflora* 'Southern Charm') ranged from 26-87% but neither use of surfactant, auxin rate, nor season impacted rooting percentage (**Table 1**).

Use of surfactant, auxin rate, or season had no effect on stomatal conductance. The three-way interaction between surfactant concentration, auxin rate, and season was significant for root number. Fall cuttings treated with a 1,500 ppm foliar IBA plus 0.85 ppm Regulaid® resulted in greater root numbers than spring cuttings treated with less than 1,500 ppm IBA with or without Regulaid®, or fall cuttings treated with a 2,500 ppm basal quick dip, a foliar application of 1,000 ppm or less without Regulaid® or fall cuttings receiving a foliar application of 0.85 ppm Regulaid®. Auxin rate impacted the average length of the three longest roots (**Table 1**). Cuttings treated with a 1,000 ppm foliar application of IBA had greater root lengths compared to cuttings treated with the 2,500 ppm IBA quick dip or cuttings receiving a 0 ppm foliar application of IBA. The interaction between auxin rate and season was significant for shoot height (**Table 2**). For spring cuttings, a 2,500 basal quick dip led to greater shoot length than cuttings receiving a foliar spray of either a 0, 500 ppm or 1,000 ppm IBA (**Fig. 1**). However, for fall cuttings, a 1,500 ppm IBA applied foliarlly led to greater shoot length than cuttings treated with 0, 500, or 2,500 ppm solution applied as a basal quick dip. Auxin rate and season impacted net photosynthesis rate (A). Cuttings treated with foliar applications of 1,500 ppm IBA had greater photosynthetic rates compared to cuttings treated with 0 ppm IBA. Cuttings taken during the spring season had greater photosynthetic rates than cuttings taken in the fall.

Table 1. Influence of surfactant, auxin and season on rooting in Teddy Bear® Southern magnolia (*Magnolia grandiflora* 'Southern Charm').

Regulaid	Auxin (ppm)	Season	Rooting (%)	Root (no.)	Avg. Length of three longest roots (cm)	Root Quality Rating[z]
			Least squares means for main effects			
0 ppm			28%	1.1 b[z]	7.6 a	2.3 b
0.85 ppm			28%	1.4 a	7.6 a	2.7 a
	Foliar IBA 0		50%	0.9 C	6.4 C	2.0 C
	Foliar IBA 500		43%	1.1 BC	7.8 ABC	2.3 BC
	Foliar IBA 1,000		62%	1.2 B	8.9 A	2.4 BC
	Foliar IBA 1,500		70%	1.7 A	8.3 AB	3.2 A
	Basal dip 2,500		56%	1.5 A	6.5 BC	2.9 AB
		Spring	54%	1.1 B	7.6 a	2.3 b
		Fall	59%	1.5 A	7.6 a	2.8 a
Regulaid	Auxin (ppm)	Season	Least squares means grouped by surfactant concentration within auxin rate by season			
	Foliar IBA 0			0 G	8.4	1.6
	Foliar IBA 500		33%	0.4 FG	8.5	1.6
0 ppm	Foliar IBA 1000		60%	0.8 EFG	8.2	2.1
	Foliar IBA 1500		53%	1.6 ABCDE	8.9	2.6
	Basal dip 2,500	Spring	60%	1.7 ABCD	6.2	3.1
	Foliar IBA 0		73%	0.9 DEFG	5.3	2
	Foliar IBA 500		26%	0.3 FG	7	1.7
0.85 ppm	Foliar IBA 1000		80%	1.0 CDEFG	9.7	2.2
	Foliar IBA 1500		73%	1.7 ABC	8	3.1
	Foliar IBA 0		40%	1.3 BCDEF	6.4	2.3
	Foliar IBA 500		60%	1.2 BDCEFG	7.3	2.4
0 ppm	Foliar IBA 1000		46%	1.2 BCDEFG	8.6	2.3
	Foliar IBA 1500		66%	1.6 ABCD	7.4	3
	Basal dip 2,500	Fall	53%	1.3 BCDEF	6.7	2.4
	Foliar IBA 0		53%	0.9 EFG	6.7	2.1
	Foliar IBA 500		60%	1.8 AB	8.2	3.2
0.85 ppm	Foliar IBA 1000		60%	1.6 ABCD	8.7	3
	Foliar IBA 1500		87%	2.0 A	8.9	3.9
Significance of treatment factors						
Surfactant			NS	0.006	NS	0.0145
Auxin Rate			NS	<0.0001	0.0018	<0.0001
Season			NS	<0.0001	NS	0.0045
Regulaid * Auxin Rate			NS	NS	NS	NS
Auxin Rate * Season			NS	<0.0001	NS	NS
Regulaid * Season			NS	NS	NS	NS
Auxin Rate * Regulaid *Season			NS	0.0026	NS	NS

[z]Root Quality (1-5, with 1 = callused without roots and 5 = ≥ 10 roots); [y]means followed by the same lower-case or upper-case letter for either main or interaction effects were not significantly different using the Holm-Simulated method for multiple comparisons ($\alpha = 0.05$), otherwise, the treatment means are presented without letter groupings for informational purposes. NS = not significant.

Table 2. Influence of surfactant, auxin and season on shoot growth, photosynthesis and stomatal conductance in Teddy Bear® Southern magnolia (*M. grandiflora* 'Southern Charm').

Regulaid	Auxin (ppm)	Season	Shoot Height (cm)	Net Photosynthesis (A) (μmol·m^{-2}·s^{-1})	Stomatal Conductance (gs_w) (mol·m^{-2}·s^{-1})
			Least squares means for main effects		
0 ppm			1.1 b[z]	3.6 a	0.1 a
0.85 ppm			2.5 a	4.3 a	0.1 a
	Foliar IBA 0		0.4 C	3.2 B	0.1 A
	Foliar IBA 500		1.4 BC	4.0 AB	0.1 A
	Foliar IBA 1,000		2.6 AB	4.2 AB	0.1 A
	Foliar IBA 1,500		3.4 A	4.7 A	0.13 A
	Basal dip 2,500		1.1 BC	3.6 AB	0.1 A
		Spring	0.66 B	6.0 a	0.1 a
		Fall	2.9 A	1.9 b	0.1 a
Regulaid	Auxin (ppm)	Season	Least squares means grouped by surfactant concentration within auxin rate by season		
0 ppm	Foliar IBA 0	Spring	0	5.1	0.10
	Foliar IBA 500		0	7.4	0.12
	Foliar IBA 1000		0	6.2	0.08
	Foliar IBA 1500		0.6	6.2	0.11
	Basal dip 2,500		2.0	7.4	0.12
0.85 ppm	Foliar IBA 0		0.7	4.9	0.07
	Foliar IBA 500		0	5.6	0.06
	Foliar IBA 1000		0.9	5.6	0.06
	Foliar IBA 1500		1.8	7.7	0.10
0 ppm	Foliar IBA 0	Fall	0	1.3	0.08
	Foliar IBA 500		0.9	1.1	0.07
	Foliar IBA 1000		4.5	2.1	0.14
	Foliar IBA 1500		3.9	2.2	0.14
	Basal dip 2,500		0.3	1.0	0.06
0.85 ppm	Foliar IBA 0		1.5	1.3	0.08
	Foliar IBA 500		4.0	3.0	0.17
	Foliar IBA 1000		4.6	2.6	0.14
	Foliar IBA 1500		6.6	2.9	0.15
Significance of treatment factors					
Surfactant			0.0013	NS	NS
Auxin Rate			<0.0001	0.048	NS
Season			<0.0001	<0.0001	NS
Regulaid * Auxin Rate			NS	NS	NS
Auxin Rate * Season			<0.0001	NS	NS
Regulaid * Season			NS	NS	NS
Auxin Rate * Regulaid *Season			NS	NS	NS

[z]means followed by the same lower-case or upper-case letter for either main or interaction effects were not significantly different using the Holm-Simulated method for multiple comparisons (α = 0.05), otherwise, the treatment means are presented without letter groupings for informational purposes. NS = not significant.

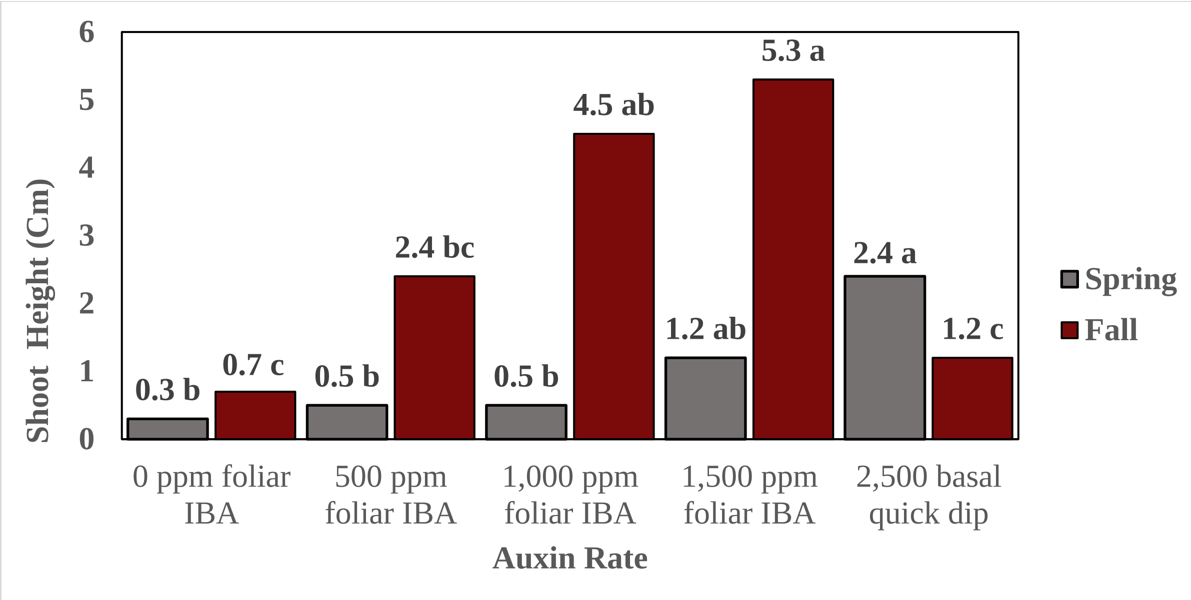

Figure 1. Shoot length of *Magnolia grandiflora* 'Southern Charm' as influenced by auxin rate and season.

DISCUSSION

The best rooting response for *M. grandiflora* 'Southern Charm' was obtained using a foliar spray of 1,500 ppm IBA or a basal quick dip of 2,500 ppm compared to foliar applications of lower concentrations. When taking cuttings in the fall, as recommended in the literature, a foliar application of 1,500 ppm IBA resulted in greater root numbers than cuttings treated with a 2,500 ppm basal quick dip. The season that cuttings were taken had a significant impact on root number, root quality ratings, and shoot growth; however, fall cuttings had lower net photosynthesis compared to spring cuttings. Previous research into photosynthetic rate during adventitious root formation suggested that the mass flow of hormones from newly formed roots were the principal contributing factor to photosynthetic rate (Svenson et al., 1995). It has further been theorized there is a correlation between a cutting's photosynthetic rate and the ability of the root initial to produce endogenous hormones (i.e., cytokinins) prior to the root elongating and penetrating the cutting surface (periderm) (Svenson et al., 1995). Physiologically, seasonal variability in endogenous hormone concentration (i.e., higher hormone concentrations in spring compared to fall) could potentially explain differences in photosynthetic rates seen in this study.

For shoot growth, fall cuttings treated with a 1,500 ppm foliar application of IBA resulted in longer shoot lengths than the 2,500 ppm IBA applied as a basal quick dip or a foliar application of 0 ppm IBA or 500 ppm IBA; however, spring cuttings treated with a 2,500 ppm basal quick dip had greater shoot lengths than cuttings treated with a 1,000 ppm foliar application of IBA or less. Nursery owners utilizing foliar auxin applications have reported slowed vegetative growth in several woody species compared to using a basal quick dip. Our results suggest that no slowed vegetative growth occurred when utilizing a foliar application of IBA compared to a basal quick dip as shoot growth was similar between a 1,500 ppm foliar application of IBA and a 2,500 ppm basal quick dip.

Our results from this trial and similar trials on foliar applications of auxin suggests that benefits of foliar applications are species dependent (Blythe et al., 2004; Bowden et al., 2021). For propagation of *Magnolia grandiflora* species and cultivars, the literature recommends higher hormone rates (5,000 to 10,000 ppm) (Dirr and Heuser, 2006). Our results suggest that sufficient auxin was absorbed from foliar applications and translocated to the site of root initiation so that root response is comparable to a basal quick dip for 'Southern Charm' magnolia. By using a foliar application of 1,500 ppm IBA on a crop of 'Southern Charm' magnolia, growers can eliminate the use of a basal quick dip for propagation of this species.

Using current methods, propagators handle a cutting multiple times before the flat even enters the propagation house, which results in higher labor costs for the producer. In addition, having several people treating and sticking cuttings can result in a wide variability in rooting, which can lead to production issues further down the process. In situations where foliar auxin applications yield equal or better results compared to traditional quick-dips, propagators only need to handle the cutting once while a licensed applicator can treat cuttings using a backpack sprayer. This helps reduce the potential for wide rooting variability. Additionally, by eliminating the need to basally treat cuttings with a stock IBA solution, the propagator can also remove the potential cross-contamination that can occur from potential pathogens that may be present on cutting stems and leaves - or from prolonged exposure to storage prior to use – when using traditional quick-dips.

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