Influence of Propagation Environment on Rooting of Sparkleberry (*Vaccinium arboreum*) Stem Cuttings©

Andrew B. Baker, James D. Spiers and Glenn B. Fain
Dept. of Horticulture, Auburn University, 101 Funchess Hall, Auburn, Alabama 36849, USA
Email: jds0017@auburn.edu

Eugene K. Blythe
Mississippi State University, Coastal Research and Extension Center, South Mississippi Branch Experiment Station, P.O. Box 193, Poplarville, Mississippi 39470, USA

Trials were conducted to determine whether propagation environment and/or substrate would encourage adventitious root formation of juvenile sparkleberry (*Vaccinium arboreum*) cuttings. The first experiment was designed in a 3×2 factorial to test the effects of three substrates (100% perlite, 2:1 perlite/ peat, 1:1 perlite/ peat) and two different environments ("mist tent" and "sweat tent") on rooting of softwood cuttings. The second experiment was designed in a 2×2 factorial to test substrate (Faford® 3B mix and a 2:1 peat/perlite mix) and ± wounding on rooting of hardwood cuttings in two separate environments (mist tent and sweat tent). Due to the low number of cuttings that rooted in all experiments, there were no significant effects of treatments on any of the parameters measured. Previous research indicates that the time of year cuttings are collected is a determining factor for successful vegetative propagation of sparkleberry.

INTRODUCTION

Sparkleberry (*Vaccinium arboreum*) has many potential uses in the landscape and fruit industry. Its uses in the commercial industry have been limited thus far by one crucial factor: *V. arboreum* is extremely difficult to vegetatively propagate. Sparkleberry has many unique landscape features including shade tolerance, exfoliating bark, berries for attracting wildlife, and great fall color. Sparkleberry also has many great features as a potential rootstock for cultivated blueberry species including, increased pH tolerance, drought resistance, and a single trunk growth habit for mechanical harvest. A successful method of asexual propagation is essential to future use in either industry.

Thus far, there has been very little research on sparkleberry propagation. Reese (1992) tested a range of auxin concentrations and found no effect on rooting. Stockton (1976) tested four levels of K-IBA on *V. arboreum* softwood cuttings with concentrations up to 20,000 ppm. No differences in rooting percentages were observed between any of the treatments. Bowerman (2012) tested a range of K-IBA concentrations on softwood, hardwood, and semi-hardwood cuttings. K-IBA concentrations up to 7,500 ppm were tested. Source and type of cutting were evaluated. Both factors were found to make a difference in rooting percentage. The highest rooting percentage (43%) occurred with the use of softwood terminal cuttings collected from water sprouts off of mature plants at the Robert Trent Jones Golf Course.

The objectives of this study were to determine the effects of substrate and environment on rooting percentages of *V. arboreum*.

MATERIALS AND METHODS

Both studies were conducted at the Paterson Greenhouse Complex, Auburn University, Auburn, Alabama. Two environments were evaluated in the two studies. The two environments were a mist tent" and a sweat tent". Rooting response (rooted or unrooted) was recorded for all cuttings, with a cutting considered rooted when any sign of adventitious roots were seen emerging from the stem. All cuttings were trimmed to 10-14 cm long.

Both environments tested were comprised of ½-in. PVC frames covered with white polyethylene film. The mist tents sat on top of expanded metal frames that were left uncovered at the base for drainage, while the sweat tents were completely enclosed by white polyethylene film. The mist tent was misted for 4 s every 10 min. The sweat tent was misted for 60 s at 8 AM and again at 1 PM.

Experiment 1

Experiment 1 was initiated on 9 Sept. 2012 using softwood cuttings. Terminal and sub-terminal cuttings were collected from Stone County, Mississippi (Lat. 30°80' N, Long. 89°17' W, U.S.D.A. Hardiness Zone 8b). Cuttings taken were juvenile cuttings arising from latent buds on mature plants that had been cut back to approximately 1 m in height in Feb 2012. This study was designed as a 3×2 complete factorial to test the effects of three substrates [100% perlite, perlite and peat (2:1, v/v), perlite and peat (1:1, v/v)] in two different environments ("mist tent" and "sweat tent"). The experimental design was a split plot design with environment as a main plot factor and substrate as a sub plot factor. There were four replications for each environment, and eight replications for each substrate. Each substrate contained two sub-samples, with six cuttings/ sub-sample. The mean day temperature in the mist tent was 18°C (65°F) ± 2.7°C (5°F). The mean night temperature was 16°C (62°F) ± 2.7°C (5°F). The mean RH was 97%. The mean day temperature in the sweat tent was 22°C (73°F) ± 5°C (9°F). The mean night temperature was 18°C (65°F) ± 2.7°C (5°F). The mean RH was 99%. Experiment 1 was terminated on 20 Dec. 2012. Additional data collected include number of cuttings that formed a callus, callus caliper (mm), number primary roots, and root length (cm).

Experiment 2

The second experiment was initiated on 28 Feb. 28, 2013 using hardwood cuttings arranged in a completely randomized design. Sub-terminal cuttings were collected from Robert Trent Jones Golf Course in Opelika, Alabama (lat. 32°69' N, long. 85°44' W, U.S.D.A. Hardiness Zone 8a). Cuttings were taken from water sprouts on mature plants. The experiment was designed in a 2×2 factorial to test substrate (Faford® 3B mix [Sun Gro Horticulture Ltd., Agawam, Massachusetts] and a perlite and peat (2:1, v/v) mix) and ± wounding in two separate environments (mist tent and sweat tent). There were 20 replications per treatment, with each cutting considered a replication. The mean day temperature in the mist tent was 24°C (76°F) ± 5°C (9°F). The mean night temperature was 22°C (72°F) ± 2.7°C (5°F). The mean RH was 92%. The mean day temperature in the sweat tent was 28°C (83°F) ± 8.3°C (15°F). The mean night temperature was 23°C (74°F) ± 4.4°C (8°F). The mean RH was 99%. Study 2 was terminated on 31 May 2013. Additional data collected include stem caliper, number of cuttings that formed a callus, callus caliper (mm), number primary roots, and root length (cm), number new leaves, number new shoots, and shoot length (cm).

Data was analyzed using generalized linear models with the GLIMMIX procedure of SAS (version 9.3; SAS Institute Inc., Cary, North Carolina). Rooting and callusing was analyzed using the binomial distribution and a log link function, count data was analyzed using the negative binomial distribution and a log link function, and measurement data was analyzed using the normal distribution and the identity function.

RESULTS AND DISCUSSION

Rooting percentages ranged from 2-8.3% in Experiment 1 (Table 1). Due to the low number of cuttings that rooted, there were no significant effects of substrate, environment, or substrate*environment on rooting, number of roots, or total root length. Though callus percentage ranged from 29.1-54.1 among treatments, there were no effects of treatments on callus or callus caliper. Though Bowerman (2012) observed the greatest rooting percentages with softwood cuttings (~40%), the cuttings were collected in May in that study while the cuttings for this experiment were collected in September. Thus the propagation, e.g., air temperature and day length, were much different than those experienced in this experiment. The time of year cuttings are collected, and/or the propagation environment of those cuttings may be important factors that contributed to the differences observed in rooting percentages in the present study compared to the study conducted on softwood cuttings by Bowerman et al. (2012).

Experiment 2 was designed to test wounding and substrate on rooting percentages of juvenile hardwood sparkleberry cuttings in two different propagation environments (mist tent and sweat tent). Very few cuttings rooted and there were no effects of substrate, wounding, or substrate*wounding on any of the data collected (Tables 2 and 3). Rooting percentages ranged from 0-7.5% in the mist tent environment (Table 2) and the sweat tent environment (Table 3). Though not significant due to the very low number of cuttings that rooted, percent rooting tended to be slightly higher in the perlite and peat (2:1, v/v) mix in both environments, perhaps due to increased water drainage. Previous research has demonstrated that hardwood cuttings of sparkleberry are very difficult to root. Bowerman (2012) observed the lowest rooting percentages when using hardwood cuttings compared to softwood and semi-hardwood. Rooting percentages in that study ranged from 0.7-10.6% for subterminal hardwood cuttings compared to 34.6-38.6% for softwood cuttings (Bowerman, 2012).

Substrates were included in the experiments presented to allow for differences in water-holding capacities in case water availability was an issue for root formation. However, substrate, wounding, and environment did not affect rooting percentages, as very few cuttings rooted in any of the experiments. The percentage of cuttings with callus formation ranged from 29-54% on softwood cuttings in Experiment 1 (Table 1) compared to 0-20% on hardwood cuttings in Experiment 2 (Tables 2 and 3). The higher percent callus formation observed on softwood cuttings may be indicative of rooting potential. Previous research has shown very little success with the vegetative propagation of sparkleberry cuttings to date. Most of the previous research conducted resulted in many treatments that do not affect rooting percentages, with the most significant finding reported by Bowerman (2012) that softwood cuttings collected in May resulted in 43.3% rooting. Thus far, rooting hormone treatments have proven to be ineffective for enhancing adventitious root formation of sparkleberry softwood, semi-hardwood, and hardwood cuttings (Bowerman, 2012; Reese, 1992; Stockton, 1976). Abscorbic acid appears to play a key role in root formation (Tyburski et al., 2006), and pre-treatment of Japanese stewartia (*Stewartia pseudocamellia*) with 0.1 M ascorbic acid prior to dipping cuttings in low IBA concentrations enhanced rooting percentages (Struve and Lagrimini, 1999). Research is currently being conducted to test the effectiveness of abscorbic acid ± K-IBA to enhance rooting percentages of softwood sparkleberry cuttings.

Literature Cited

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Table 1. Effect of environment and substrate on rooting of juvenile sparkleberry (*Vaccinium arboreum*) softwood cuttings.[z]

Substrate	Environment	Callus (%)	Callus caliper (mm)	Rooting (%)	Roots (no.)	Root length (cm)
1:1 perlite/peat	Mist[y]	37.5	5.6	4.1	1.5	3.9
2:1 perlite/peat	Mist	33.3	4.3	2.0	1.0	3.5
100% perlite	Mist	29.1	3.8	4.1	1.0	0.4
1:1 perlite/peat	Sweat[x]	54.1	8.2	6.3	2.7	7.8
2:1 perlite/peat	Sweat	47.9	9.0	8.3	1.5	3.2
100% perlite	Sweat	43.7	5.4	6.3	3.3	3.7
Significance[w]		NS	NS	NS	NS	NS

[z] Softwood cuttings taken 20 Sept., 2012 from Stone County, MS. Juvenile cuttings were taken from latent buds on mature plants that had been cut back to approximately 1 m in height in Feb. 2012.

[y] "Mist tents" were covered with white polyethylene plastic and placed into intermittent mist 4 s every 20 min.

[x] "Sweat tents" were constructed using white polyethylene plastic on all sides with mist provided for 60 s at 8 am and 1 pm.

[w] Nonsignificant (NS).

Table 2. Effect of wounding and substrate type on rooting, callus formation, and new growth of sparkleberry (*Vaccinium arboreum*) hardwood cuttings in "mist tent"[z] environment.[y]

Substrate	Wound	Stem caliper (mm)	Rooting (%)	Roots (no.)	Root length (cm)	Callus (%)	Callus caliper (mm)	New shoots (no.)	Shoot length (cm)	New leaves (no.)
Faford® 3B mix	N	4.2	0	*	*	0	*	*	*	*
Faford® 3B mix	Y	4.5	2.5	3	2.0	0	*	8.0	5.0	63
Perlite:peat (2:1, v/v)	N	4.4	7.5	1	1.7	10	2.5	3.6	3.0	25
Perlite:peat (2:1, v/v)	Y	4.6	5.0	2	2.5	5	4.3	5.5	4.8	28
Significance[x]	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

[z] "Mist tents" were covered with white polyethylene plastic and placed into intermittent mist 4 s every 20 min.

[y] Hardwood cuttings were taken 28 Feb., 2012 from Robert Trent Jones Golf Course in Opelika, AL, cuttings taken from water sprouts of mature plants.

[x] Nonsignificant (NS).

Table 3. Effect of wounding and substrate type on rooting, callus formation, and new growth of sparkleberry (*Vaccinium arboreum*) hardwood cuttings in "sweat tent"[z] environment.[y]

Substrate	Wound	Stem caliper (mm)	Rooting (%)	Roots (no.)	Root length (cm)	Callus (%)	Callus caliper (mm)	New shoots (no.)	Shoot length (cm)	New leaves (no.)
Faford® 3B mix	N	3.8	0	-	-	0	-	0	-	0
Faford® 3B mix	Y	3.8	0	-	-	0	-	0	-	0
Perlite:peat (2:1, v/v)	N	4.4	7.5	5.0	4.2	5	4.2	3.6	3.6	30
Perlite:peat (2:1, v/v)	Y	4.6	2.5	4.0	5.0	20	4.7	5.0	4.0	30
Significance[x]	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

[z] "Sweat tents" were constructed using white polyethylene plastic on all sides with mist provided for 60 s at 8 am and 1 pm.

[y] Hardwood cuttings were taken on 28 Feb. 2012 from Robert Trent Jones Golf Course in Opelika, AL. Cuttings were taken from water sprouts of mature plants.

[x] Nonsignificant (NS).