Southern Region Small Fruit Consortium

Final Report Research

Title: Evaluation of a Rooting Protocol for Hardwood Cuttings of Muscadine Grapes

Grant Code: SRSFC Project # 2021-R11

Grant Period: March 1, 2021-February 28, 2022

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Public Abstract

Rooting hardwood cuttings from muscadine (*Vitis rotundifolia* Michx.) vines has traditionally been considered an exceptionally difficult task. Many previous studies observed almost no root formation, leading to a general consensus that muscadines should either be propagated by softwood cuttings or vegetative layering. However, the University of Arkansas System Division of Agriculture Fruit Breeding Program has been using a hardwood rooting protocol for muscadines with moderate success for the past ten years. The application of this protocol to meet the modest propagation needs of the breeding program has significantly shortened the time required to advance selections. The goal of this research was to more adequately describe the factors affecting the rooting ability of hardwood muscadine cuttings. This research investigated the effects of cultivar, bottom heat, cold storage, vineyard location, and cutting collection date on the outcome of muscadine hardwood cuttings. The study was conducted during the dormant seasons of 2019-2020 and 2020-2021, and an overall rooting percentage of 16% was observed. There were multiple higher-order interactions affecting rooting efficacy. Cuttings taken in November generally rooted at higher rates, although interactions with vineyard location and cultivar played a significant role in those results. The Ocilla, GA location performed

exceptionally well in November with rooting percentages of over 40%. The effects of supplying bottom heat and/or a cold storage treatment on rooting success declined as the dormant season progressed. Other variables such as increased cutting length and diameter were associated with increased rooting success. Ultimately, this research shows that institutions with modest muscadine propagation needs can successfully propagate plants from hardwood cuttings.

Introduction

Muscadines (*Vitis rotundifolia* Michx.) are a species of grape native to the southeastern U.S. This specialty crop is recognized for its disease resistance and thick-skinned, large-seeded berries. Unlike bunch grape species such as *V. labrusca* and *V. vinifera* that are relatively easy to propagate, muscadines are notoriously difficult to propagate by hardwood cuttings (Himelrick, 2001). Therefore, most muscadine breeders, nurseries, and germplasm repositories propagate muscadines by layering or rooting softwood cuttings under mist. In a breeding program, the selection of muscadine seedlings occurs at the end of the growing season in September. A reliance on softwood cuttings requires the postponement of propagation of selected seedlings until the next growing season, generally in June or July. This propagation schedule falls into the busiest time of the year for fruit breeding programs and delays the establishment of plots and evaluation of new selections by a full season. This yearlong delay adds considerable time to the already lengthy process of releasing a variety. The development of a reliable protocol for muscadine propagation by hardwood cuttings would allow propagation work to be conducted after the conclusion of the growing season at a time when work in the field is beginning to slow and would increase the speed of cultivar development.

The literature on propagation of muscadines is sparse and most of the work conducted in this area is very old. Recommendations by Niven (1918) highlight the variability of rooting success for muscadine cultivars and suggest that 8-10% rooting success would be "excellent work." Cuttings were placed in well-prepared soil with only 1-2 inches of wood above the soil surface. Propagation by hardwood cuttings was only recommended if one was willing to get minimal success (Niven, 1918). Later studies at the University of Georgia found rooting percentages from 0 to 3% depending on cultivar; however, there was a noted difference in success when cuttings were taken later in the year. The rooting medium used was fine sand mulched with composted leaves, using wood of various ages. Rooting percentages were improved, but still not commercially viable, when cuttings were taken in November compared to August. These failures were attributed to poor callusing (Woodroof, 1935). Both of the aforementioned studies overwintered propagules in outdoor nursery beds. The application of bottom heat to cuttings in greenhouses greatly increased the success of cuttings taken in November, with rooting percentages as high as 70% (Newman, 1907). More recent investigations into rooting in greenhouses with bottom heat had mixed results. Hardwood cuttings from 'Hunt' were taken in Georgia from November to February on four different dates and treated with and without bottom heat. Rooting of 1-2% of cuttings with bottom heat was observed and no roots were found on cuttings without bottom heat (Goode et al., 1982). Another study found that muscadines rooted readily when hardwood cuttings were held at 4 °C for 60-90 days before potting (Whatley, 1974).

Despite all the literature claiming that muscadine propagation with hardwood cuttings is ineffective, the University of Arkansas System Division of Agriculture Fruit Research Station (UA-FRS) has been using hardwood cuttings to meet its modest muscadine propagation needs for the past 15 years. The methods used at the Fruit Research Station were adapted from the protocol used for hardwood propagation of bunch grapes. Using these methods, success rates are highly variable, depending on both cultivar and year, ranging from 10-70%. The goal of this SRSFC-funded study was to test the efficacy of the FRS propagation protocol and the impact of multiple factors frequently mentioned in previous studies that may affect rooting success.

Objectives

- 1. Determine the efficacy of a clonal propagation protocol for hardwood cuttings used by the University of Arkansas breeding program.
- 2. Assess the impact of collection date, location, cold storage, and bottom heating on the outcome of hardwood cuttings taken from muscadine grapes.

Materials and Methods

There were five factors tested in this study:

- Location- Vineyards in Clarksville, AR, Fayetteville, AR, and Ocilla, GA.
- Cultivar- 'Fry', 'Carlos', and 'Supreme.'
- Collection Date- Cuttings were taken once at the beginning of November, December, January, and February during the winters of 2019/2020 and 2020/2021.
- Storage- Half of all cuttings were given a month-long cold storage treatment at 4 °C prior to the rooting treatment.
- Bottom Heat- Half of all cuttings were given continuous bottom heat at 26 °C.

Cuttings were collected from mature vines at each vineyard location. Cuttings were approximately 15-20 cm long and 5-10 mm wide, with a minimum of three nodes. The rooting containers used for this study were SureRoots® Deep Cell 50-cell plug trays (T.O. Plastics, Clearwater, MN) with 12.7 cm deep cells. Trays were cut into 10-cell experimental units to facilitate replication and randomization within the study. The rooting media was 100% perlite. Cuttings were dipped in 0.1% Indole-3-butyric acid powder (Bonide Products Inc., Oriskany, NY) before being inserted into the rooting media such that one node was fully submerged in the rooting media and a second node was level with the surface of the rooting media. One cutting was planted into each cell.

Ambient temperatures in the greenhouse were maintained between 18-24 °C through the course of the study. Two greenhouse benches measuring 1.5 m x 3.0 m were used as mist beds. A 1.9 cm in-line sprinkler valve (Rain Bird Corp., Azusa, CA) was connected to a standard hose valve at native city water pressure. A Galcon 8056S AC-6S (Galcon USA LTD., Simi Valley, CA) programmable irrigation controller was wired to the valve. The valve was programmed to run the

mist system for 15 s every 10 min with an irrigation window of 6:00 AM to 6:00 PM. The irrigation line was 0.64 cm in diameter and suspended approximately 0.61 m above the mist benches. Three Coolnet Pro foggers (Netafim Irrigation Inc., Fresno, CA) were spaced evenly lengthwise across each bench. These foggers were a four-nozzle system and each nozzle flowed at 7.6 L.h-1. In addition, an internal check valve inside each fogger ensured that shut off happened quickly after valve closure to maintain a consistent 15 s mist interval. The media was also hand watered to field capacity approximately twice a week during the study as well as immediately after cuttings were placed in the media to ensure adequate moisture availability.

Data on cutting length, cutting diameter, and number nodes per cutting were also collected 90 days after the cuttings were treated with rooting hormone and placed in perlite media under a mist system in a heated greenhouse.

The proportion of cuttings that rooted in each experimental unit was analyzed using PROC GLIMMIX in SAS 9.4 (SAS Institute Inc., Cary, NC) as a mixed model with five fixed effect treatments, two random effects, and a negative binomial distribution. Bottom heat was the main plot effect and the other four factors (collection date, cold storage, cultivar, and location) were completely randomized within the split plot. Block and year were analyzed as random effects. In order to interpret higher-order interactions, the results of mean separations tests were sliced by location when applicable. Mean separations were performed with Tukey's Honest Significant Difference. Graphical representations of data were constructed using the ggplot2 package in RStudio (Wickham, 2016).

Results and Discussion

The percentage of cuttings that formed roots for the two years of this study were 17.1% and 15.0%, respectively, for an overall rooting percentage of 16.0%. There were four higher order interactions that significantly affected the proportion of hardwood muscadine cuttings that rooted: Location x Cultivar x Date, Location x Heat x Date, Heat x Storage x Date, and Location x Cultivar x Storage.

Generally, cuttings taken in November outperformed cuttings taken at other dates. This trend is most notable at the Ocilla, GA vineyard, where rooting percentages were over 40% (Figure 1). We hypothesize that this is due to the incomplete dormancy of those vines. Located in southern Georgia, Ocilla had not experienced its first frost by the time cuttings were taken in November. However, cuttings taken in November also performed well in the other study locations. Supplying bottom heat early in the dormant season appears to have helped stimulate rooting, but did not seem to have an effect on cuttings taken in January or February. In addition, increased cutting diameter and cutting length positively affected rooting success.

Conclusion

This research provides showed that rooting muscadines from hardwood cuttings is a more effective method of propagation than many previous studies have concluded. The complex experimental design of the study allowed for the higher-order interactions affecting the rooting success of muscadine hardwood cuttings to be examined. Cuttings taken from Georgia in early

November formed roots more than 40% of the time, perhaps indicating that an incomplete transition to dormancy may have played a role in the success of this study and others. When cuttings from November were analyzed alone with location and cultivar considered random effects, applying a cold storage treatment reduced rooting percentages from 27% to 19% and the effect of bottom heat was non-significant. Increased cutting length and cutting diameter were significantly correlated with increased rooting success while an increased number of nodes was not. Thus, While the rooting percentages reported in this study may not allow for commercially successful propagation of muscadines by hardwood cuttings, breeding programs or germplasm repositories with modest needs may find that transitioning to an off-season propagation protocol may save time and money.

Impact Statement

This collaborative research from the University of Arkansas System Division of Agriculture (UA System) and University of Georgia (UGA) generated data that can be used to develop recommendations for hardwood propagation of muscadine grapes to shorten breeding cycles and increase flexibility for breeding programs and germplasm repositories. The data from this study has been presented at the National ASHS meeting and published in the Fall 2021 issue of Small Fruit News. This project will also be a chapter of Kenneth Buck's UA Horticulture Department Master's Thesis and an expanded manuscript based on our results is currently under review in HortScience.

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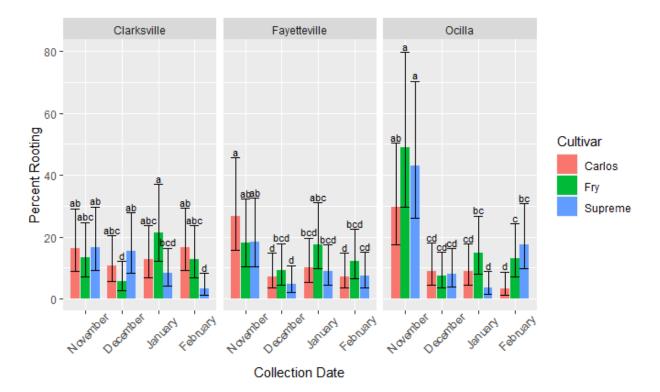


Figure 1. Percent of cuttings successfully rooted from 'Carlos', 'Fry', and 'Supreme' vines collected in early November, December, January, and February from vineyards in Clarksville, AR, Fayetteville, AR, and Ocilla, GA. Because of a significant three-way interaction, data is presented separately for each location. Means with different letter(s) in each panel are significantly different (a = 0.05) according to Tukey's honestly significant difference.



Figure 2. Muscadine vines that have been successfully rooted from hardwood cuttings.