## CONCLUSION

The response of the 'Marina' to air-layering was good, with the first rooted layers harvested about 90 days after placement. The response to the hormone dilutions was rapid and acceptable, with the 5000 and 7500 ppm dilutions consistently producing moderate and abundant callus as early as 38 days post air-layer placement. Stem diameters between 7.0 and 12.0 mm had the best results, with the most abundant callus formation and earliest root formation. The 4.5- to 6.5-mm diameter stems had quick callus formation, but were more inclined to break or otherwise fail. The results of soil inoculation of the moist air-layer material appears equivocal to date, with no consistent trends noted. The 'Marina's had a very gratifying response to long-day lighting, producing exuberant new growth in January, very close to the level of growth seen in the summer. This may indicate a possibility that the 'Marina' might also favorably respond to long-day lighting with callus and root formation at a similar rate and quality as that produced during the summer months. This might indicate that air-layering propagation of *A.* 'Marina' may be possible on a year-round basis, without loss of productivity during the shorter, colder days of winter, if they were long-day lighted and provided adequate warmth during the winter.

# Improving Handling and Rooting of *Thunbergia alata*©

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## INTRODUCTION

*Thunbergia alata*, common name "black-eyed Susan vine," is an increasingly popular ornamental vine named for the black eyes of its miniature flowers. *Thunbergia alata* is commonly propagated by seed and stem-and-two-leaf nodal cuttings. Large-scale cutting production of *T. alata* is performed in Costa Rica by Ball Flora Plant. Many of the cuttings experience leaf disintegration and literal melting usually within the first week of planting, resulting in failure to root and death. A study was launched to examine factors affecting the rooting of *T. alata* Sunny™ Lemon Star black-eyed Susan PPAF vine cuttings.

## MATERIALS AND METHODS

Unless otherwise noted, no cuttings of *T. alata* were treated with rooting hormones in the following experiments.

**Carbohydrates and Rooting.** One hundred-fifty *T. alata* Sunny™ Lemon Star black-eyed Susan cuttings were separated into basal (nodes 1 and 2) and nonbasal (nodes 3 and 4) treatment groups to examine the effect of internodal cutting position on rooting. Hamilton et al. (2002) found that leaf size of *Coleus* cuttings greatly influenced rooting quality. Lemon Star basal cuttings often have larger leaves than nonbasal cuttings. Cuttings were planted in sterile Oasis® cubes, placed in a misting house with bottom heat under intermittent mist at 4 sec mist every 4 min, and propagation survival was recorded after 3 weeks.

**Respiration and Temperature.** The effect of temperature on respiration rate of *T. alata* Sunny™ Lemon Star black-eyed Susan was examined using a static

system, with cuttings held at 2.2 ºC, 7.2 ºC, and 20 ºC. Pint Mason jars were sealed with 4 cuttings per jar and placed for 1 h at 20 ºC and 2 h at 2.2 ºC and 7.2 ºC. Levels of $CO_2$ were measured with a gas chromatograph fitted with a CTR-1 column (Alltech). Jars containing cuttings were filled with distilled water and weighed to determine actual headspace. Data was recorded as mg $CO_2$ per $kg \cdot h^{-1}$.

**Temperature and Shipping Potential.** Sixty-eight *T. alata* Sunny™ Lemon Star black-eyed Susan cuttings were placed in permeable vegetable bags at 2.2 ºC and 20 ºC for 3 days to assess the effect of abusive temperatures during shipment from Costa Rica to the United States. Cuttings were planted in sterile Oasis cubes and placed in a misting house with bottom heat under intermittent mist at 4 sec mist every 4 min. The number of cuttings rooting after 1 week was determined.

**Misting Interval and Rooting.** The effect of intermittent mist duration on cutting survival was examined. Treatments consisted of misting *T. alata* Sunny™ Lemon Star black-eyed Susan cuttings for 4 sec every 4 min for 3 weeks, 4 sec mist every 4 min for 1 week then 4 sec mist every 8 min for 2 weeks, and 4 sec mist every 8 min for 3 weeks. Cuttings were planted in sterile Oasis cubes with bottom heat, and survival was recorded after 3 weeks.

**Hormones and Rooting.** Cuttings of *T. alata* Sunny™ Lemon Star black-eyed Susan were treated with Dip 'N Grow® (1% IBA and 0.5% NAA) at 1 : 5, 1 : 10, and 1 : 20 dilutions; Rootone® (0.2% NAA) dipping powder; or tap water. All cuttings were planted in sterile Oasis cubes and placed under intermittent mist at an interval of 4 sec every 8 min. Bottom heat was applied to the rooting bed. Propagation survival was recorded after 3 weeks.

## RESULTS

**Carbohydrates and Rooting.** Nonbasal cuttings rooted better than basal cuttings (chi-square analysis p = 0.000) (Fig. 1). This was surprising as research had indicated that the larger store of carbohydrates in large-leaf cuttings (basal) could

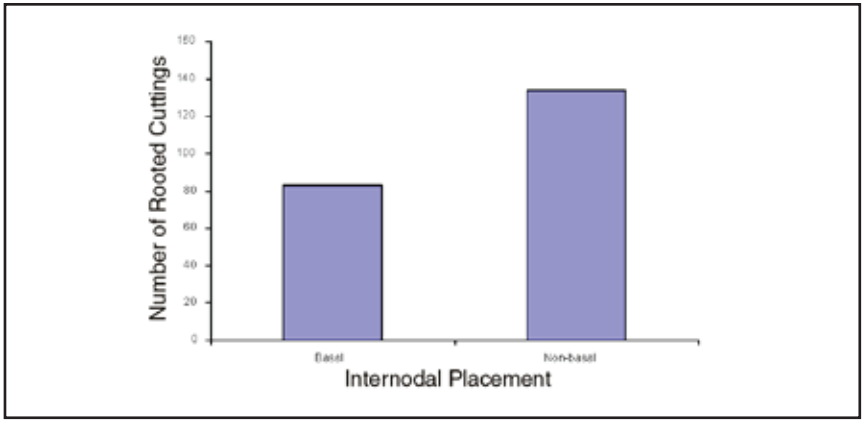


**Figure 1.** The effect of internodal placement on cutting survival of *T. alata*, Sunny™ Lemon Star black-eyed Susan. Nonbasal cuttings taken at nodes 3 and 4 rooted at a greater rate (chi-square analysis P = 0.000) than basal cuttings taken at nodes 1 and 2. Cuttings were placed in a rooting bed with bottom heat for 3 weeks and misted every 4 min for 4 sec. One hundred-fifty cuttings were assessed from each location.

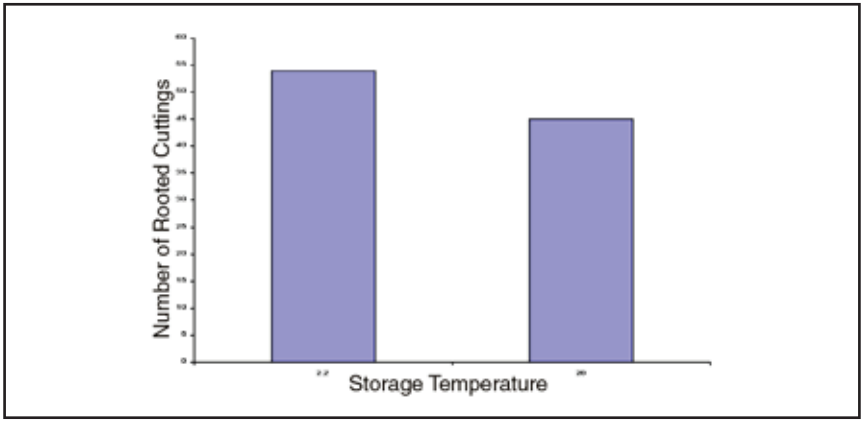


**Figure 2.** The effect of storage temperature on the rooting of cuttings of *Thunbergia alata* Sunny™ Lemon Star black-eyed Susan. Cuttings were stored for 3 days at 2.2 or 20 ºC before propagation. The values represent the results after 3 weeks with intermittent misting occurring every 4 min for 4 sec. Sixty-eight cuttings were stored at each temperature.

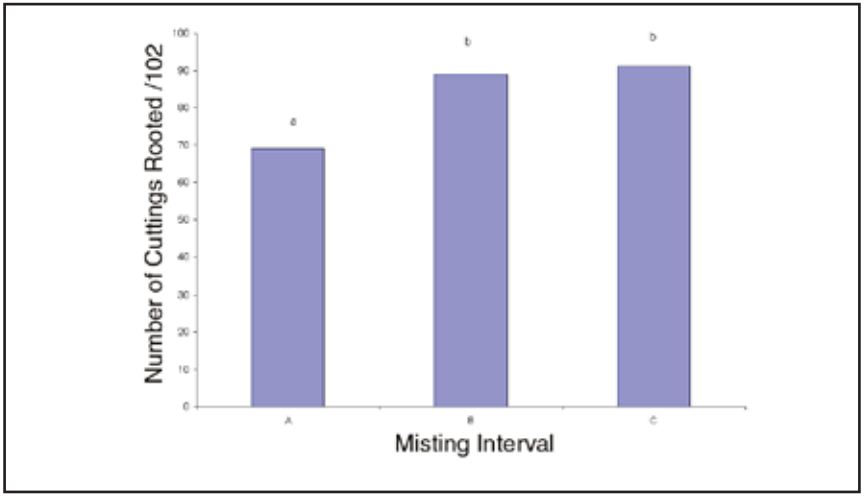


**Figure 3.** The effect of misting duration on cutting survival of *Thunbergia alata,* Sunny™ Lemon Star black-eyed Susan. These results represent data collected after 3 weeks in mist propagation at variable rates of misting duration. Means with the same letter are not significantly different at the 95% confidence level according to Tukey's Mean Separation Test.

A = 4 sec mist every 4 min for 3 weeks.

B = 4 sec mist every 4 min for 1 week then 4 sec mist every 8 min for 2 weeks.

C = 4 sec mist every 8 min for 3 weeks.

enable these cuttings to root easier (Veierskov, 1988). With *T. alata* 'Lemon Star', it was observed that basal cuttings with larger leaves had a higher incidence of leaf disintegration, which could have led to the lower incidence of rooting.

**Respiration and Temperature.** There was a dramatic increase in respiration rate as temperature increased (Fig. 2). The respiration rate of cuttings at 20 ºC (591 mg $CO_2$ per kg·h$^{-1}$) was 11.5 times higher than that of cuttings at 2.2 ºC (23 mg $CO_2$ per kg·h$^{-1}$). The respiration rate of cuttings held at 7.2 ºC was intermediate at 58 mg $CO_2$ per kg·h$^{-1}$. These results indicate that uneven and/or high temperatures during shipment could deplete the carbohydrate reserves of cuttings, reducing their ability to successfully root.

**Temperature and Shipping Potential.** There was no difference in rooting between cuttings held at 2.2 ºC or 20 ºC for 3 days, though cuttings held at 2.2 ºC tended to root to a greater extent than those held at 20 ºC (chi-square analysis $p = 0.083$). Replication of this trial also demonstrated a slight tendency for higher rooting but statistically no difference between the rooting of each group.

**Misting Interval and Rooting.** Cuttings subjected to 4 sec mist every 4 min for 1 week then 4 sec mist every 8 min for 2 weeks, or 4 sec mist every 8 min for 3 weeks had a higher rooting percentage than those cuttings subjected to 4 sec mist every 4 min for 3 weeks (Tukey's Mean Separation Test) (Fig. 3). When cuttings were misted at the shorter frequency (every 4 min), the leaves appeared waterlogged and chlorotic and the leaves disintegrated more rapidly than those misted every 8 min.

**Hormones and Rooting.** The Dip 'N Grow dilutions appeared phytotoxic to the cuttings, causing yellowing and melting of the leaves and eventual cutting death (Fig. 4). Interestingly, though, cuttings treated with Rootone powder rooted better than the cuttings treated with the Dip 'N Grow dilutions; the Rootone-treated cuttings rooted to the same extent as the cuttings treated with tap water.

## DISCUSSION

There is very little published literature regarding the propagation of *T. alata*, so the rooting problem had to be examined from a practical standpoint. *Thunbergia alata* cuttings tended to root best when taken from the nonbasal nodal position (nodes 3 and 4). This was attributed to less surface area for leaching in the mist and thus less leaf disintegration. Also, it is known that auxin is produced at the shoot tips, so perhaps increased auxin levels in nonbasal cuttings was promoting rooting.

Temperature clearly affected the respiration rate of Sunny™ Lemon Star black-eyed Susan, though it did not appear to affect the rooting of cuttings stored at 2.2 or 20 ºC. *Thunbergia alata* cuttings are typically packaged with an ice pack, but the ice packs have been observed to be completely melted upon arrival. Thus, the cuttings may be subjected to relatively high temperatures during transit. The small tendency for cuttings stored at cold temperature to root better does not seem to justify the potential added cost of refrigeration. In addition, it has yet to be determined if *T. alata* is chilling sensitive.

Lemon Star cuttings rooted better when the mist interval was increased from 4 to 8 min. Cuttings were not as waterlogged or chlorotic when the interval was increased, and this was correlated with rooting success. As cuttings with larger

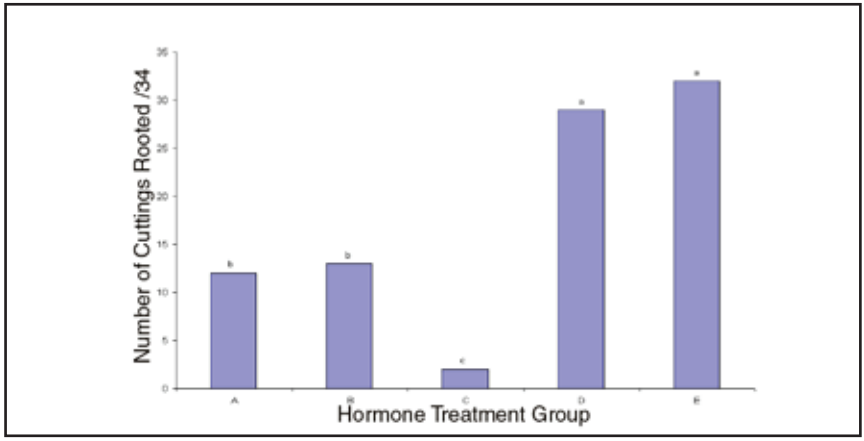


**Figure 4.** Effect of rooting hormones on the rooting of cuttings of *Thunbergia alata* Sunny™ Lemon Star black-eyed Susan cuttings. Cuttings were treated with specified rooting hormones, and rooted cuttings were counted after 3 weeks with bottom heat and intermittent mist at 4 min every 8 sec. Means with the same letter are not significantly different at the 95% confidence level according to Tukey's Mean Separation Test.

A = Dip 'N Grow® 1% IBA, 0.5% NAA at 1 : 5 dilution ratio.

B = Dip 'N Grow 1% IBA, 0.5% NAA at 1 : 10 dilution ratio.

C = Dip 'N Grow 1% IBA, 0.5% NAA at 1 : 20 dilution ratio.

D = Rootone® 0.2%NAA.

E = Tap water.

leaves tended to not root as well as those with smaller leaves, increasing the misting interval may promote higher rooting of larger-leaf cuttings.

Ball Flora Plant does not recommend the use of hormones when propagating *T. alata* cuttings. The study comparing Dip 'N Grow to Rootone or tap water indicates that auxin does not increase rooting, which supports Ball's recommendation.

Research on *T. alata* propagation will be conducted this spring analyzing the effects of plant growth regulators on stock plant cutting production and the effects of water quality and droplet size on rooting.

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