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Flowering *Psychotria ligustrifolia* (Northrop) Millsp. (Rubiaceae).

# Cutting propagation of 4 Florida native taxa of wild coffee (*Psychotria* spp.)

Teagan H Young, Sandra B Wilson, Mack Thetford, and James Colee

## ABSTRACT

Wild coffee (*Psychotria nervosa* Sw. [Rubiaceae]), softleaf wild coffee (*Psychotria tenuifolia* Sw.), and Bahama wild coffee (*Psychotria ligustrifolia* (Northrop) Millsp.) are evergreen shrubs with attractive foliage, fragrant white flowers, and colorful fruit. A cutting propagation study was conducted to determine the effects of auxin concentration on rooting of these 3 species, and a dwarf form, *Psychotria nervosa* 'Little Psycho'. We treated semi-hardwood stem cuttings of each taxa with one of 3 talc indole-3-butyric acid (IBA) concentrations (0, 8000, and 16,000 mg/kg) and evaluated for rooting responses. Cuttings for all taxa rooted by 8 wk under mist. Root system quality values increased for *P. nervosa* 'Little Psycho' and *P. tenuifolia* with auxin application, while root system quality values for *P. ligustrifolia* and *P. nervosa* were similar among auxin treatments. Percentage of cuttings with roots was similar among taxa and increased with IBA at 8000 and 16,000 mg/kg compared to the control. Both root length and root number were influenced by auxin application, and the effects of auxin differed among taxa. Except for *P. nervosa*, all taxa treated with IBA produced roots that were longer than control cuttings. Auxin application increased root number compared to the control for *P. ligustrifolia*, *P. nervosa*, and *P. nervosa* 'Little Psycho'. *Psychotria tenuifolia* had the greatest number of cuttings with 25 or more roots, but root number did not differ regardless of auxin concentration. Results confirm that native Florida *Psychotria* taxa are ideal candidates for vegetative propagation by semi-hardwood stem cuttings.

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## KEY WORDS

Bahama coffee, dwarf wild coffee, softleaf wild coffee, Rubiaceae, vegetative propagation, auxin treatment

## NOMENCLATURE

USDA NRCS (2022)  
Wunderlin and others (2022)

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Photos by Sandra Wilson and Teagan Young

## CONVERSIONS

$(^{\circ}\text{C} \times 1.8) + 32 = ^{\circ}\text{F}$

25.4 mm = 1 in

2.54 cm = 1 in

1 m = 3.3 ft

0.43 kg = 1 lb

mg/kg = parts per million (ppm)

**N**ative plants have been historically overlooked in their value to urban landscapes and gardens despite their importance in attracting pollinators and providing wildlife habitat (Burghardt and others 2009; Pardee and Philpott 2014; Kalamán and others 2022a; Kalamán and others 2022b). The appeal of using native plants in landscapes has increased, however, and homeowners are willing to pay more for a residential landscape that includes well-designed and strategically placed native plants (Helfand and others 2006; Gillis and Swim 2020). Underutilization of native plants can be attributed to inefficient or unknown propagation systems, insufficient marketing and promotion, and limited availability in consumer markets (Dumroese and others 2009; Wilde and others 2015; Wilson 2020). Moreover, the majority of native species are not available in the market. For example, White and others (2018) identified slightly more than 800 active native plant vendors selling approximately 26% (~6500 species) of all US native flora. Propagation knowledge is key to increasing the diversity within a native plant palette. While propagation protocols have been developed for a number of underutilized native species (Thetford and others 2018; Trigiano and others 2021; Campbell-Martínez and others 2022; Smith and others 2022), propagation knowledge for the majority of US native species is yet to be discovered.

The fourth largest angiosperm family, Rubiaceae, consists of both economically and ornamentally important genera including *Psychotria*. The largest genus within Rubiaceae, *Psychotria* is represented by many understory plants that protect against soil erosion, attract pollinators such as birds and butterflies, and have medicinal value (Francis 2004; Coelho and others 2013; Calixto and others 2016). Additionally, *Psychotria* have ornamental value as they can be used as hedges, borders, and mass plantings. Three *Psychotria* species are native to Florida: wild coffee (*Psychotria nervosa* Sw.), softleaf wild coffee (*Psychotria tenuifolia* Sw.), and Bahama coffee (*Psychotria ligustrifolia*

(Northrop) Millsp.) (Gann and others 2022; Wunderlin and others 2022). Although *P. ligustrifolia* is listed as critically imperiled in south Florida, the conservation status of both *P. nervosa* and *P. tenuifolia* is listed as secure (Gann and others 2022). Characteristics common among these taxa include leaves that are simple, oppositely arranged, with entire margins and prominent veins; inflorescences that are cymose with flowers that are white and fragrant (Figure 1A); fruits that are 2-seeded, longitudinally ribbed drupes with a fleshy pericarp that turns red in the fall (Figure 1B; Burch and others 1975); and fibrous root systems (Figure 1C). Collectively, wild coffees can be used as hedges, borders, and mass plantings (Burch and others 1975), with flowering in the summer and fruiting in the fall. Native to Florida's peninsula, *P. nervosa* and *P. tenuifolia* occur in mesic and rockland hammocks (FNAI 2010), are 1.2 to 2.4 m tall, grow in USDA hardiness zones 9a–11, and are moderately salt tolerant (USDA 2012) (Table 1). *Psychotria nervosa* 'Little Psycho' is a dwarf cultivar first identified by Brightman Logan in a west-central Florida hammock and is now in commercial micropropagation (AgriStarts 2022) (Table 1). It has a shorter more horizontal form, rarely reaches a maximum of 0.3 to 0.9 m in height, and has internodes shorter than the other taxa. *Psychotria ligustrifolia* is a rare species native to mainland and Florida Keys rockland hammocks (FNAI 2010). It is shorter than the other taxa (0.9–1.2 m in height), less cold tolerant (cold hardiness of 9b–11) (USDA 2012), more tolerant of full sun conditions, and possesses low salt tolerance (Table 1). These *Psychotria* taxa can be readily distinguished from one another by their leaves and fruit (Figure 1D). Leaves of *P. nervosa* and *P. nervosa* 'Little Psycho' are shiny green and very conspicuously veined, and they have drupes that are oblong and red. *Psychotria nervosa* 'Little Psycho' has larger drupes compared to the other 3 taxa. Leaves of *P. tenuifolia* are dull and have a more blueish-green cast. Drupes are smaller than 5.8 × 5.5 mm and are orange to red in color. Leaves of *P. ligustrifolia* are



290 *Psychotria nervosa* drupes.



*Psychotria tenuifolia* drupes.

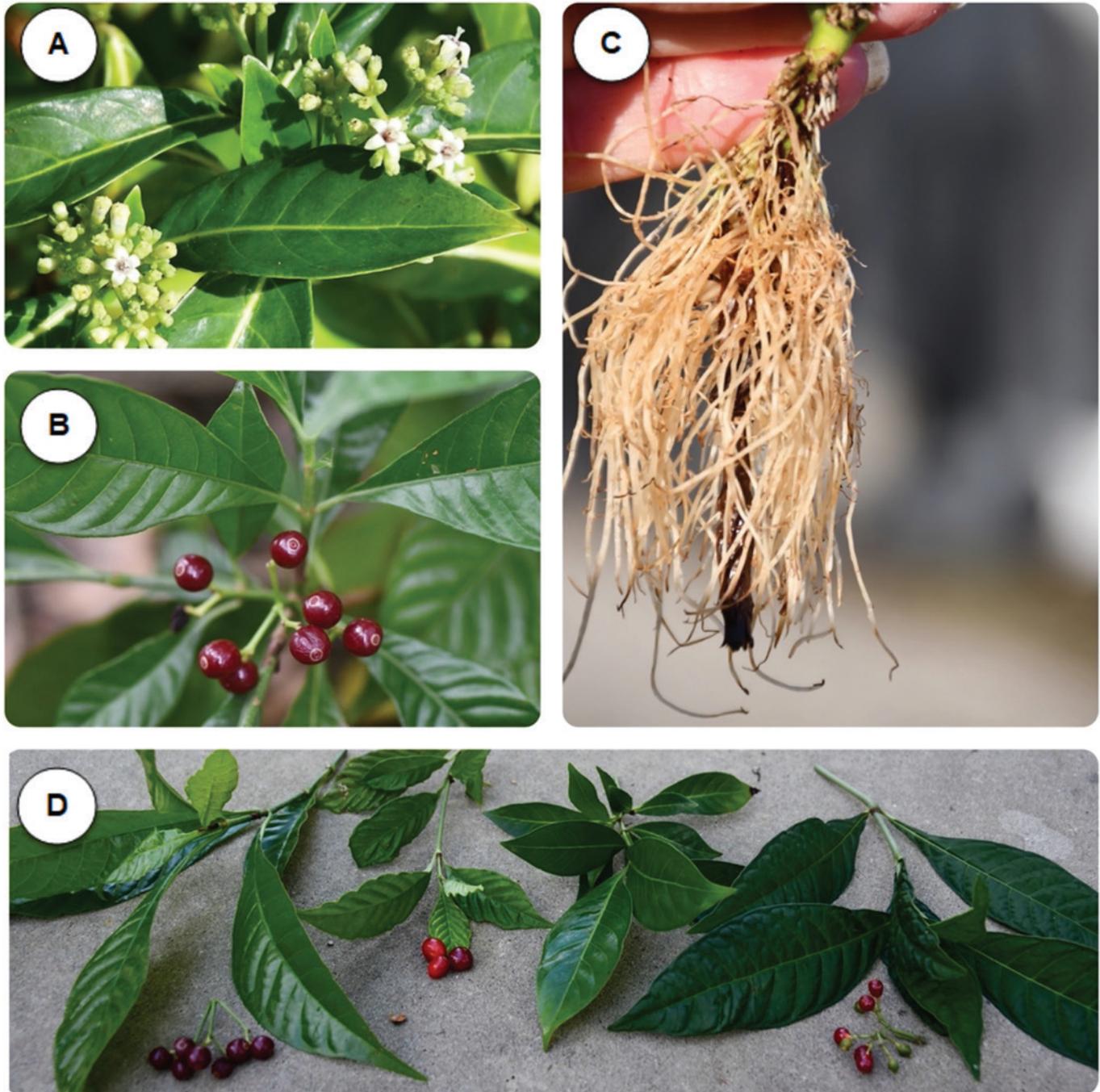


Figure 1. Typical flowering, fruiting, and foliage of *Psychotria* spp. with prominently veined, elliptical leaves (A–B); white, cymose, terminal inflorescences (A) that are followed by 2-seeded, ovoid drupes eventually turning red in color (B). Cuttings form well-developed root systems in 8 wk (C). Side-by-side comparison of 4 taxa, from left to right: *Psychotria nervosa*, *Psychotria nervosa* 'Little Psycho', *Psychotria ligustrifolia*, and *Psychotria tenuifolia* (D).

smaller than *P. nervosa* and *P. tenuifolia* and possess a lighter colored midrib with less vein prominence. Their fruit size is larger than *P. tenuifolia*, measuring  $6.4 \times 6.1$  mm (Figure 1D).

Propagation information is limited for *Psychotria*. Seeds are reported to have physiological dormancy and require long periods of time to germinate (Garwood 1983; Baskin and others 2004; Francis 2004; Athugala and others 2016). In a preliminary study, Pereira and Wilson found spring and summer to

be more ideal than fall or winter for seed germination of *P. ligustrifolia*, *P. nervosa*, and *P. tenuifolia*, yet germination was inconsistent ranging from 9% to 80% over 5 mo (unpublished data). Moreover, seeds are difficult to collect in large volumes and are thought to lose viability quickly (Godts 2022), thus propagation by stem cuttings may be a practical alternative.

However, cutting propagation protocols for Florida native *Psychotria* are largely lacking within the literature. A literature

TABLE 1

Native distribution, ecosystem, description, and attracted pollinators of 4 *Psychotria* taxa evaluated for cutting propagation.

| Scientific name                           | Common name          | Cold hardiness zone <sup>z</sup> | Florida distribution <sup>y</sup> | Florida ecosystem <sup>y</sup>             | Description <sup>x</sup>   | Pollinator / wildlife use                       |
|---|----------------------|----------------------------------|-----------------------------------|--|--|---|
| <i>Psychotria ligustrifolia</i>           | Bahama wild coffee   | 9b to 11                         | Southern peninsula                | Rockland hammocks and pine rocklands       | 0.9–1.2 m (3–4 ft) tall, 0.9–1.2 m (3–4 ft) wide, green foliage, 6.4 × 6.1 mm (0.25 × 0.24 in) showy red drupes        | Birds, bees, wasps, flies                       |
| <i>Psychotria nervosa</i>                 | Wild coffee          | 9a to 11                         | Peninsula                         | Hammocks                                   | 1.2–2.4 m (4–8 ft) tall, 1.2–2.4 m (4–8 ft) wide, glossy green foliage, 8.1 × 6.9 mm (0.32 × 0.27 in) showy red drupes | Birds, butterflies, bees, wasps, flies, beetles |
| <i>Psychotria nervosa</i> ‘Little Psycho’ | Dwarf wild coffee    | 9a to 11                         | Cultivated                        | Found in a hammock in central west Florida | 0.3–0.9 m (12–36 in) tall, glossy green foliage, 8.8 × 7.7 mm (0.34 × 0.30 in) showy red drupes larger than the parent | Bees, butterflies                               |
| <i>Psychotria tenuifolia</i>              | Softleaf wild coffee | 9a to 11                         | Central and southern peninsula    | Hammocks                                   | 1.2–2.4 m (4–8 ft) tall, 1.2–2.4 m (4–8 ft) wide, dull green foliage, 5.8 × 5.5 mm (0.23 × 0.22 in) showy red drupes   | Bees, butterflies, birds                        |

<sup>z</sup> USDA 2012.

<sup>y</sup> Wunderlin and Hansen 2011.

<sup>x</sup> Plant size descriptors were based on observations of plants in north-central Florida. For drupe size, 10 drupes were measured and presented as a mean.

review of these taxa yielded a single study. Denny and West (2008) reported a positive correlation between auxin concentration, percentage of rooting, and cutting quality of *P. nervosa*, although specific information is lacking as this was an abstract that was never published into a peer-reviewed article. Still, it indicates cutting propagation is feasible for some Florida native *Psychotria*. Cutting propagation of other *Psychotria* has been demonstrated within cell culture propagation systems, indicating species within the genus can be propagated from stem cuttings (Kerber and others 2001; Magedans and others 2019). Moreover, other species in Rubiaceae can be propagated from stem cuttings and benefit from auxin application. For example, Haldu (*Adina cordifolia* Roxb.) had increased rooting with high auxin concentrations (20,000 mg/kg IBA) compared to lower auxin concentrations for 1 to 2 cm diameter cuttings (Yadav and others 2018). Similarly, the application of 3000 or 5000 mg/kg IBA improved rooting for Chinese emmenopterys (*Emmenopterys henryi* Oliv. [Rubiaceae]) compared to other auxin concentrations for semi-hardwood terminal cuttings taken during the summer (Aiello and Dillard 2013).

The overall goal of this study is to widen the use of *Psychotria* taxa in landscapes by developing practical methods for commercial cutting propagation. Specific objectives were to determine the effects of taxa and auxin concentration on optimal rooting responses of *Psychotria*.

## MATERIALS AND METHODS

292 Cuttings of *P. ligustrifolia*, *P. nervosa*, *P. nervosa* ‘Little Psycho,’ and *P. tenuifolia* located on the University of Florida

Campus were taken on the morning of 23 October 2020. A total of 54 semi-hardwood cuttings from each taxa were selected that had a terminal bud, a minimum of 5 nodes, was pest- and disease-free, and lacked fruits. Cutting lengths were 8 to 9.5 cm for *P. ligustrifolia* and *tenuifolia*, 5 to 8.5 cm for *P. nervosa*, and 4.5 to 9.5 cm for *P. nervosa* ‘Little Psycho’. We removed the basal leaves, then dipped the 1.3 cm basal portion of each stem in tap water prior to commercial talc rooting hormone that contained either 0, 8000, or 16,000 mg/kg (ppm) indole-3-butyric acid (IBA; Hormex, Mainland, Pennsylvania). We dipped the control treatments (0 mg/kg IBA) into water only. After treatment application, cuttings were stuck into 6-cell trays (width 3.8 cm × length 3.8 cm × depth 5.8 cm) (T.O. Plastics, Clearwater, Minnesota) filled with Metro-Mix 852 (6:3:1 bark:Canadian peat:perlite) (Sun Gro Horticulture, Agawam, Massachusetts). We hand-misted the cuttings to keep them from wilting until transferred to the misthouse. Each auxin treatment was applied to 6 cuttings replicated 3 times in a randomized complete block design. Overhead mist was provided every 5 min at 5 sec time intervals for 8 wk by a Sterling 12 irrigation controller (Superior Controls, Seabrook, New Hampshire) and upright mister nozzles (Senninger Irrigation, Clermont, Florida) spaced 76.2 cm apart along a single greenhouse bench. The average, maximum, and minimum temperatures in the mist house were 25.1 °C, 36.4 °C, and 13.7 °C, respectively (HOBO Pendant MX Water Temperature Data Logger; Onset Computer Corporation, Bourne, Massachusetts). Cuttings were checked weekly for root emergence, pest interference, and foliage loss.



Teagan Young collecting cutting samples from *Psychotria tenuifolia*.

At the end of the experiment (after 8 wk), we evaluated rooting by gently pulling cuttings out of individual cells to determine a root system quality value using a scale from 0 to 4, with 0 = dead cuttings; 1 = alive cuttings with no roots; 2 = roots forming but do not hold medium; 3 = rootball partially holds plug medium; and 4 = fully formed rootball entirely holding the medium. We gently removed the substrate from the roots to collect additional rooting data. Rooting percentages were calculated on all cuttings. Root system characteristics were evaluated for cuttings with roots (cuttings with root quality ratings of 1–4) and included root length (mean of the 2 longest roots) and root number with a maximum of 25 roots counted.

### Statistical Analysis

Data were analyzed in a two-step process; the first part was to test for interactions between taxa and auxin, then based on those results nonparametric tests were used to determine statistical differences in auxin, either across or for each taxa. This two-step method was used because of non-normality, right censoring, and boundary issues that made a traditional method inaccurate. For this analysis to be completed, subsamples were assumed independent. Root system quality value, rooting percentage, root length, and root number data were analyzed using a linear mixed model with JMP v. 16 (SAS Institute, Cary, North Carolina). Taxa, auxin, and the taxa  $\times$  auxin interaction were treated as fixed effects while block was treated as random effect. If the type III test for interaction were significant ( $P \leq 0.05$ ), then we used the non-parametric Steel-Dwass multiple comparison procedure to compare auxin treatments by taxa. Root number data were analyzed by taxa using the Kruskal-Wallis non-parametric ANOVA followed by the non-parametric Steel-Dwass multiple comparison procedure to account for non-normality and censored data (due to root counts stopping at 25).

The root system quality value reflects the combined effects of rooting percentage, root length, and root number of all living cuttings. This overall assessment value (scale of 1–4 where 1 = no roots, 2 = roots that did not hold media, 3 = roots that partially held media, and 4 = roots that completely held media) was influenced by auxin application and these effects differed among taxa (taxa  $\times$  auxin interaction,  $P = 0.0201$ ) (Table 2). Root quality values for *P. ligustrifolia* and *P. nervosa* (2.2–2.9 and 1.9–2.4, respectively) were similar among auxin treatments, whereas root quality values for *P. nervosa* ‘Little Psycho’ and *P. tenuifolia* were significantly improved with an auxin application (Table 3). As such, *P. nervosa* ‘Little Psycho’ had nearly twice the root quality score when treated with auxin compared to the control (1.6 compared to 2.9 to 3.6) (Table 3). *Psychotria tenuifolia* treated with 8000 mg/kg IBA also had higher root quality (3.6) than the control cuttings (2.7), and cuttings treated with 16,000 mg/kg IBA had a similar root quality (3.1) compared to cuttings treated with 0 or 16,000 mg/kg IBA (Table 3).

We measured a significant auxin effect for percent rooting ( $P < 0.0001$ ). However, rooting percentage did not statistically differ by taxa ( $P = 0.0860$ ) (rooting percentages were 87.0%, 87.0%, 85.2%, and 98.1% for *P. ligustrifolia*, *P. nervosa*, *P. nervosa* ‘Little Psycho’, and *P. tenuifolia*, respectively) nor was there

TABLE 2

*Linear mixed model significance for root system quality, percent rooting, root length, and root number based on taxa, auxin concentration, and the interaction between taxa and auxin concentration.*

| Effect  | Num df | Den df | F value | P value |
|---|--------|--------|---------|---------|
| <b>Root system quality (scale of 1–4)</b>       |        |        |         |         |
| Taxa  | 3      | 198.5  | 7.5494  | <0.0001 |
| Auxin concentration                             | 2      | 198.5  | 19.4220 | <0.0001 |
| Taxa $\times$ auxin                             | 6      | 198.4  | 2.5736  | 0.0201  |
| <b>Root percentage</b>                          |        |        |         |         |
| Taxa  | 3      | 198.1  | 2.2292  | 0.0860  |
| Auxin concentration                             | 2      | 198.1  | 10.6760 | <0.0001 |
| Taxa $\times$ auxin                             | 6      | 198.1  | 1.2450  | 0.2849  |
| <b>Root length (average of 2 longest roots)</b> |        |        |         |         |
| Taxa  | 3      | 176.1  | 12.0370 | <0.0001 |
| Auxin concentration                             | 2      | 176.1  | 16.1550 | <0.0001 |
| Taxa $\times$ auxin                             | 6      | 176.0  | 2.5054  | 0.0237  |
| <b>Root number</b>                              |        |        |         |         |
| Taxa  | 3      | 175.6  | 4.3660  | 0.0054  |
| Auxin concentration                             | 2      | 176.3  | 55.2760 | <0.0001 |
| Taxa $\times$ auxin                             | 6      | 175.8  | 2.4243  | 0.0282  |

TABLE 3

Mean root system quality rating and root length  $\pm$  standard error of the mean of 4 *Psychotria taxa* subjected to 3 auxin treatments (0, 8000, and 16,000 mg/kg [ppm] indole-3-butyric acid [IBA]).

| Taxa <sup>z</sup>                         | IBA (mg/kg)  | Root system quality <sup>y</sup><br>(1 to 4 scale) | n <sup>x</sup> | Rooting length <sup>w</sup> (mm) | n <sup>x</sup> |
|---|--------------|--|----------------|----------------------------------|----------------|
| <i>Psychotria ligustrifolia</i>           |              |  |                |                                  |                |
|   | 0 mg/kg      | 2.22 $\pm$ 0.24 a                                  | 18             | 23.21 $\pm$ 3.58 b               | 14             |
|   | 8000 mg/kg   | 2.83 $\pm$ 0.25 a                                  | 18             | 43.06 $\pm$ 4.83 a               | 16             |
|   | 16,000 mg/kg | 2.88 $\pm$ 0.18 a                                  | 18             | 33.82 $\pm$ 5.75 ab              | 17             |
| <i>Psychotria nervosa</i>                 |              |  |                |                                  |                |
|   | 0 mg/kg      | 1.89 $\pm$ 0.18 a                                  | 18             | 18.08 $\pm$ 4.28 a               | 13             |
|   | 8000 mg/kg   | 2.44 $\pm$ 0.23 a                                  | 18             | 27.72 $\pm$ 3.73 a               | 16             |
|   | 16,000 mg/kg | 2.44 $\pm$ 0.21 a                                  | 18             | 29.06 $\pm$ 3.52 a               | 17             |
| <i>Psychotria nervosa</i> 'Little Psycho' |              |  |                |                                  |                |
|   | 0 mg/kg      | 1.61 $\pm$ 0.12 b                                  | 18             | 10.05 $\pm$ 1.36 c               | 11             |
|   | 8000 mg/kg   | 2.89 $\pm$ 0.18 a                                  | 18             | 32.06 $\pm$ 4.53 b               | 18             |
|   | 16,000 mg/kg | 3.59 $\pm$ 0.15 a                                  | 18             | 44.21 $\pm$ 2.03 a               | 17             |
| <i>Psychotria tenuifolia</i>              |              |  |                |                                  |                |
|   | 0 mg/kg      | 2.67 $\pm$ 0.24 b                                  | 18             | 36.74 $\pm$ 4.95 b               | 17             |
|   | 8000 mg/kg   | 3.64 $\pm$ 0.18 a                                  | 14             | 55.25 $\pm$ 2.81 a               | 14             |
|   | 16,000 mg/kg | 3.06 $\pm$ 0.22 ab                                 | 18             | 43.78 $\pm$ 5.22 ab              | 18             |

<sup>z</sup> For each taxa, means within a column followed by the same letter are not significantly different according to a Steel-Dwass test at  $P \geq 0.05$ .

<sup>y</sup> Cuttings were evaluated using a visual root system quality scale from 1 to 4 with 1 = alive cuttings with no roots; 2 = roots forming but do not hold medium; 3 = rootball partially holds plug medium; and 4 = fully formed rootball entirely holding the plug medium when removed from the tray. Dead cuttings were not included in analysis.

<sup>x</sup> Total number ( $n$ ) of cuttings included in analysis are designated in columns after each measured trait.

<sup>w</sup> Rooting lengths of the 2 longest roots were averaged. Root length can only be measured for cuttings with roots, hence, green stems without roots were not included in analysis.

a taxa  $\times$  auxin interaction ( $P = 0.2849$ ) (Table 2). The percentage of cuttings with roots was greatest for cuttings treated with auxin (88 $\pm$ 4% and 88 $\pm$ 2% for 8000 and 16,000 mg/kg auxin, respectively) compared to cuttings not treated with auxin (control) (71 $\pm$ 10%) (Table 4).

Both root length and root number were influenced by auxin application, and the effects of auxin differed among taxa (taxa  $\times$  auxin interaction;  $P \leq 0.237$  and  $P \leq 0.282$ , respectively) (Table 2). For root length, cuttings of *P. ligustrifolia* and *P. tenuifolia* treated with 8000 mg/kg IBA produced roots that were approximately 2.0 and 1.5 times longer (43 and 55 mm), respectively, than control cuttings (23 and 37 mm) while cuttings treated with 16,000 mg/kg IBA had a similar root length (34 and 44 mm) compared to cuttings treated with 0 and 8000 mg/kg IBA (Table 3). For *P. nervosa*, auxin treatment did not improve root length regardless of auxin level (8 to 29 mm). For *P. nervosa* 'Little Psycho', however, auxin application did result in longer roots compared to the control, and a higher auxin concentration further increased root length. *Psychotria nervosa* 'Little Psycho' cuttings treated

with 16,000 mg/kg had the longest roots (44 mm), followed by 8000 mg/kg (32 mm), while control cuttings had the shortest roots (10 mm) (Table 3).

Root number was influenced by auxin application for 3 of the 4 taxa (Table 2). Auxin application increased root number

TABLE 4

Mean percent rooting  $\pm$  standard error based on the Agresti–Coull Percent estimates method of the combined 4 *Psychotria taxa* subjected to 3 auxin treatments (0, 8000, and 16,000 mg/kg [ppm] indole-3-butyric acid [IBA]).

| IBA (mg/kg) <sup>z</sup> | Rooting percentage (%) | n  |
|--------------------------|------------------------|----|
| 0 mg/kg                  | 70.60 $\pm$ 9.53 b     | 72 |
| 8000 mg/kg               | 87.77 $\pm$ 4.38 a     | 72 |
| 16,000 mg/kg             | 87.77 $\pm$ 2.29 a     | 72 |

Notes: Means within a column followed by the same letter are not significantly different according to a Steel-Dwass test at  $P \geq 0.05$ . Total number ( $n$ ) of cuttings included in analysis are designated in the right-hand column.

<sup>z</sup> Rooting percentage did not differ by taxa ( $P = 0.0861$ ), and no taxa  $\times$  auxin interaction was observed ( $P = 0.2849$ ).

TABLE 5

Auxin concentration treatments (0, 8000, and 16,000 mg/kg [ppm] indole-3-butyric acid [IBA]) and quantiles for root number of rooted cuttings for 4 *Psychotria* taxa.

| Taxa                                      | IBA (mg/kg)    | Number of roots <sup>z</sup> |
|---|----------------|------------------------------|
|   |                | Median (25%, 75%)            |
| <i>Psychotria ligustrifolia</i>           |                |                              |
|   | 0 mg/kg b      | 6.50 (1.75, 18.75)           |
|   | 8000 mg/kg a   | 25.00 (15.00, 25.00)         |
|   | 16,000 mg/kg a | 23.00 (11.50, 25.00)         |
| <i>Psychotria nervosa</i>                 |                |                              |
|   | 0 mg/kg b      | 5.00 (2.50, 10.50)           |
|   | 8000 mg/kg a   | 25.00 (25.00, 25.00)         |
|   | 16,000 mg/kg a | 20.00 (8.50, 25.00)          |
| <i>Psychotria nervosa</i> 'Little Psycho' |                |                              |
|   | 0 mg/kg c      | 8.00 (2.00, 12.00)           |
|   | 8000 mg/kg b   | 25.00 (22.75, 25.00)         |
|   | 16,000 mg/kg a | 25.00 (25.00, 25.00)         |
| <i>Psychotria tenuifolia</i>              |                |                              |
|   | 0 mg/kg a      | 14.00 (4.00, 25.00)          |
|   | 8000 mg/kg a   | 25.00 (25.00, 25.00)         |
|   | 16,000 mg/kg a | 25.00 (22.50, 25.00)         |

Notes: For each taxa, auxin levels followed by the same letter are not significantly different according to a Steel-Dwass test at  $P \geq 0.05$ .

<sup>z</sup>A maximum of 25 roots were counted per rooted cutting.

compared to the non-treated control for *P. ligustrifolia*, *P. nervosa*, and *P. nervosa* 'Little Psycho' whereas root number for *P. tenuifolia* did not differ regardless of auxin concentration (Table 5). For *P. ligustrifolia* and *P. nervosa* root number was similar among cuttings treated with 8000 or 16,000 mg/kg IBA, and these cuttings had more roots than the control cuttings had (Table 5). *Psychotria nervosa* 'Little Psycho' had the greatest number of roots when treated with 16,000 mg/kg followed by 8000 mg/kg, compared to the control. Quartile data for all 4 taxa revealed that 75% of the rooted cuttings had 25 roots or more when treated with either auxin concentration. Similarly, 50% of rooted cuttings had 25 roots or more with the exceptions of *P. ligustrifolia* (23 roots) and *P. nervosa* (20 roots) at 16,000 mg/kg (Table 5). For all 4 taxa, root number for control cuttings with roots within the median quartile was considerably lower (5–14 roots per cutting) compared to cuttings receiving auxin. Control cuttings within the 75% quartiles show a similar pattern of lower root numbers except for *P. tenuifolia* for which 75% of the rooted cuttings had 25 roots or more. The only taxa with 75% of rooted cuttings producing 25 or more roots for both control and auxin treated cuttings was *P. tenuifolia*.

Similar to several species in Rubiaceae, *Psychotria* taxa native to Florida can be reliably commercially propagated from stem cuttings using methods standard in the nursery industry (Ferreira and others 2014; Nery and others 2014; Yu and others 2014; Yadav and others 2018). More specifically, related ornamental shrubs such as Firebush (*Hamelia patens* Jacq.) (Davis and others 1991), Ixora (*Ixora* L.), and Gardenia (*Gardenia* Ellis.) are considered easy to propagate using stem cuttings treated with 3000 mg/kg IBA (Davies and others 2018). Depending on the species and its demand, and the size of the nursery operation, rooting percentages between 50% and 80% could be considered acceptable for native nursery production (Cartabiano and Lubell 2013; Godts 2022). While commercially acceptable rooting percentages (>70%) were obtained without the application of auxin, rooting percentage was increased approximately 1.2 times and with a positive effect on root quality, root number, and root length for cuttings treated with 8000 mg/kg IBA, but with only minimal improvement with the use of 16,000 mg/kg IBA.

This study utilized very high IBA concentrations at 8000 or 16,000 mg/kg, a level typically reserved for hard-to-root species (Davies and others 2018). High auxin concentrations have the potential to adversely affect rooting, particularly with alcohol-based solutions (Davies and others 2018), but this did not appear to be the case for *Psychotria*. Mean rooting percentages among the 4 Florida taxa were between 71% and 88% and were not that different from the 89% spring and 61% summer results obtained by Nery and others (2014) for *P. nuda* rooted with 0 to 3000 mg/kg IBA. Thus, it is possible that lower IBA concentrations of 1000 or 3000 mg/kg would also suffice for the Florida native *Psychotria* taxa, thereby reducing auxin cost.

Additional factors, such as seasonality of cutting harvests and (or) cutting maturity (flowering, fruiting), could potentially affect the rooting success of *Psychotria* (Davies and others 2018), although testing such factors was not an objective of this study. Nery and others (2014) found spring to be the most favorable season for rooting *P. nuda*. In our study, cuttings were taken in the fall at the same time some plants were fruiting. Although care was taken not to collect cuttings from fruiting branches, the presence of fruit on the plants may have affected rooting success (Davies and others 2018). It is possible Florida native *Psychotria* could also be rooted during other times of the year, which could offer a potential improvement to overall rooting percentages or result in greater improvement of root quality. Anecdotally, we successfully rooted cuttings of *P. nervosa* stuck in late spring with similar rooting performance. The similarity of fall rooting percentages of *Psychotria* taxa reported herein to the spring and summer rooting percentages of *P. nuda*, suggest a high likelihood that *P. nervosa*, *P. tenuifolia*,

and *P. ligustrifolia* can be clonally propagated year-round in Florida.

The root system quality values generated herein reflect the combined effects of rooting percentage, root length, and root number and summarize a visual assessment of the entire root system of all living cuttings. This overall assessment value is useful in summarizing the observations for the individual taxa, but if used alone it may not reflect the impacts of individual root quality measures. For example, *P. ligustrifolia* had similar root quality values reflecting cuttings with rootballs partially holding plug medium with a greater proportion of cuttings holding the plug medium. Root number data further suggest IBA at 8000 mg/kg will result in more cuttings with 25 or more roots per cuttings. *Psychotria nervosa* values reflect root systems that partially held plug medium with a proportion of the cuttings not holding plug medium. The increased root number resulting from auxin-treated cuttings was likely contributing to the increased proportion of cuttings that held the plug medium. For *P. nervosa* 'Little Psycho', very few cuttings held the medium in the absence of auxin. Application of auxin increased both root length and root number, resulting in higher quality root systems. *Psychotria tenuifolia* demonstrated the highest root system quality ratings among the non-treated control cuttings. Regardless of auxin application, most *P. tenuifolia* cuttings partially held the plug medium. However, a larger proportion of cuttings had fully formed rootballs and a larger proportion of the cuttings had 25 or more roots with auxin applied at 8000 mg/kg.

## CONCLUSIONS

*Psychotria* taxa have many positive traits such as drought tolerance, ornamental appeal, and fragrant flowers, and they provide valuable pollinator and wildlife resources that make it desirable for use in Florida landscapes. Propagation results demonstrate these taxa are relatively easy to produce in a propagation greenhouse under intermittent mist from fall stem cuttings, leading to a finished liner within 8 wk. While these taxa may be rooted from stem cuttings without auxin application, we recommend the application of IBA to improve rooting percentages and improve measures of root quality such as root number and root length. Among all 4 taxa, at least one of the measures of rooting performance improved with a talc formulation containing 8000 mg/kg IBA, but it is possible lower concentrations may also suffice. Continued advances in propagation knowledge of these taxa may be developed with the exploration of timing (seasonality) of cutting collection and small, on-site propagation trials with lower concentrations or alternative formulations of IBA. The differing results for the 2 *P. nervosa* taxa also suggest an improvement of propagation may result not only from a refinement of either auxin source or concentration but additionally by selection of desirable, easy-to-root clones.

Further development of propagation information will allow for more efficient liner production, an increase in nursery production, and potentially a widened use of *Psychotria* taxa in landscapes and gardens.

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## AUTHOR INFORMATION

**Teagan H Young**  
MS Graduate Student  
teagan.young@ufl.edu

**Sandra B Wilson**  
Professor  
sbwilson@ufl.edu

**Mack Thetford**  
Associate Professor  
thetford@ufl.edu

**James Colee**  
Consultant  
Statistical Consultant Unit  
colee@ufl.edu

Department of Environmental Horticulture  
University of Florida  
Building 550, Mehrhof Hall  
PO Box 110670  
Gainesville, FL 32611-0670

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