UF IFAS
UNIVERSITY of FLORIDA

FNGLA Endowment 2021-2022 Funding Reports

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ABOUT THE FNGLA ENDOWMENT

The Florida Nursery, Growers and Landscape Association (FNGLA) created an endowment in 2005 to address problems and questions that are important to the Florida nursery industry.

The FNGLA Endowed Research Fund (#F003129/30) provides awards up to $5,000 each to supplement and extend existing research projects. The principal balance of the endowment is more than $1.45 million, and **9 projects** were funded for a total of **$44,789** and involved **16 faculty members**.

The following priorities were determined for the selection of the 2021-2022 projects:

1. Enhance Floridians' Quality of Life
2. Improve Environmental and Resource Management (no projects this year)
3. Improve Pest Management Practices and Strategies
4. Improve Production Systems Practices and Strategies
5. Genetics and Breeding to Enhance Quantities and Diversity of Plant Material

The selection process included a review by the following FNGLA committee members:

- Ed Bravo
- Stefan Liopiros
- Mike Marshall
- Van Donnan
- David McDonald
- Sylvia Gordon
- Linda Reindl

A MESSAGE OF THANKS

To the Florida Nursery, Growers and Landscape Association:

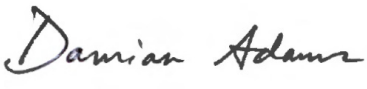
Thank you for your support. The funding your organization raised allows UF/IFAS researchers to continue their great work towards research and discovery both on campus and at our off-campus research centers (see map on the final page of this document).

We also want to thank the selection committee for the time they dedicated to this program. Your thorough review ensures the projects that receive funding are the best of the best.

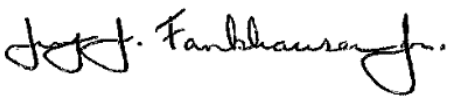
We look forward to this continued collaboration and hope you find this document helpful.

Sincerely,

Robert Gilbert
UF/IFAS Dean for Research
Director of the Florida Agricultural Experiment Station (FAES)



Damian Adams
UF/IFAS Associate Dean for Research
Associate Director of the FAES



Jerry Fankhauser
Assistant Director of the FAES

ENHANCE FLORIDIANS' QUALITY OF LIFE

This priority area is defined as:

FNGLA supports research that will improve or enhance the quality of life for Floridians.

FNGLA supported one project under this priority area, and that summary is on pages 5-10.

How does plant diversity, vegetation structure, and management contribute to ecosystem services in residential landscaping

PI: **Basil Iannone, School of Forest, Fisheries & Geomatics Sciences**

Co-PI: **Jesse Jones, School of Forest, Fisheries & Geomatics Sciences**

ABSTRACT

With over 900 new residents moving to Florida daily, it is critical to design residential landscapes that provide the benefits expected by homeowners while mitigating impacts of development on Florida's valuable environmental resources. Our project contributes to this need by determining how landscaping management practices, plant community diversity, and vegetative structure interact to affect ecosystem services of importance to urban residents (e.g., aesthetics, wildlife habitat, yard utility, cooling, maintenance costs). To understand the interacting dynamics of landscaping, maintenance practices, and ecosystem services, we administered homeowner questionnaires and performed field surveys of yards across a socioeconomic gradient in Gainesville, FL (N=90). Preliminary results indicate the relationship between a maintenance intensity index (reflecting use of irrigation, fertilizer, and pesticides) and the performance of ecosystem services important to homeowners can be summarized as everything from no relationship to strong negative relationships. In other words, the priorities of homeowners do not align with resource-intensive management typical to the existing landscaping industry and supply chains. FNGLA funding was granted to perform analyses of soil fertility that have been collected and mostly processed, but we are still awaiting finalization of total carbon and total nitrogen before pursuing more in-depth analyses of soil fertility. Moving forward, we will develop predictive models that inform the design of sustainable, cost-effective residential landscapes that deliver the benefits people want from their yards. These models will guide industry (e.g., nursery growers, landscape architects/designers/installers, maintenance professionals) and inform policy aiming to promote environmentally responsible landscaping in our State's rapidly expanding residential areas. These collective objectives directly meet FNGLA's research priority of enhancing Floridians' quality of life.

OBJECTIVES AND METHODS

1. Determine what ecosystem services homeowners prioritize from their yards and how current landscaping norms achieve those outcomes.

2. Assess components of plant biodiversity that contribute to the performance of ecosystem services in residential landscapes.

3. Create predictive models to inform the design and adoption of sustainable landscaping practices.

Methods

We recruited participants by mailing invitations to approx. 1,800 homeowners throughout Gainesville, FL requesting they complete a survey about their yard's characteristics, maintenance practices, and costs.

To focus on mature residential landscapes, we targeted neighborhoods with homes at least 30 years old. We further selected for neighborhoods that exhibited large variance in outdoor water use along a socioeconomically diverse gradient. This presumably captured a variety of landscaping strategies while controlling for differences caused by socioeconomic affluence (e.g., the "luxury effect"). From the larger pool of recruitment survey respondents, we selected a subset of participants with varied landscaping practices. The resulting study sample includes 90 yards across 12 neighborhoods ranging from conventional HOAs to underserved communities, capturing a diversity of landscaping styles typical to residential areas of the southeastern US. We collected in-depth ecological data from each of these yards quantifying plant species richness and cover, herbivory, and soil fertility (**Figure 2**). For the latter, we took eight soil cores from each yard to quantify total carbon (C), total nitrogen (N), C:N ratio, pH, and organic matter (OM), all of which are important indicators of soil fertility and health. FNGLA funding was used for part of this task. We also measured vegetation structure using LiDAR drone flights. Follow-up surveys were then administered to homeowners to assess their landscaping priorities and the benefits they receive from their yards.

Initial analyses involved development of a maintenance intensity index to assess the extent to which variation in management strategies deliver the ecosystem services important to homeowners. This index was developed via principal component analysis of water usage estimates from the H2OSAV utility database and data on a consortium of maintenance practices reported by homeowners along an ordinal scale of intensity. The main principal component axis (PC1) which reflected use of irrigation, fertilizer, and pesticides was then related to the performance of ecosystem services (assessed from both field data and Likert scale homeowner survey responses) using either Pearson or Kendall Tau correlation tests.

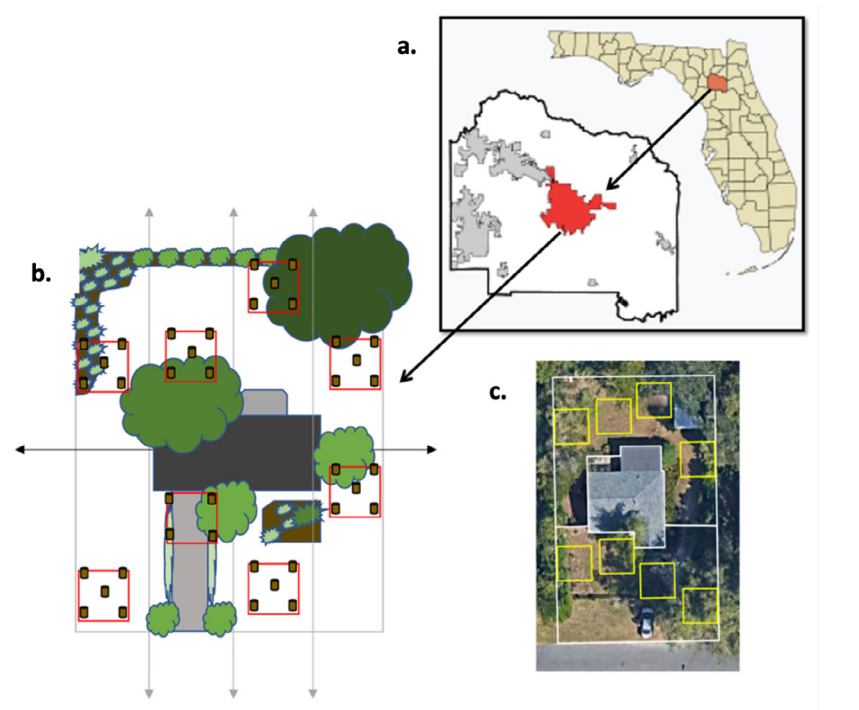


Figure 1. Map showing geographic location of study area in the city of Gainesville, Alachua County, Florida (a), diagram illustrating stratified field sample design developed to represent spatial heterogeneity of plant community typical in residential yards (b), and a study site with true location of plots overlaid (c).

Figure 2. Dr. Vitor Vasconcelos, Kacey Russo, and Olesya Malakhova performing a vegetation survey on one of the study's yards.

RESULTS

The participant-performed ranking exercise of "priorities when making landscaping decisions" is quantified by ranked position, so lower values equate to higher overall prioritization among homeowners (**Figure 3**). The ranking results panned out in some ways as expected with high ranking of *aesthetics* and *comfortable environment*, but the importance of *wildlife habitat* and *native plants*, alongside *property value* as the lowest average priority, came as a surprise.

The landscaping maintenance PCA constructed from data on management practices and outdoor water usage resulted in an ordination where PC1 explained 29.5% of the variance in the data, and this variance was related to the degree to which irrigation, fertilizer, and pesticides are used in a yard's upkeep (**Figure 4**).

The relationship between the landscaping management intensity index, i.e., household scores along PC1, and ecosystem service outcomes (assessed thru a mix of field and social data) resulted in everything from no correlation to strong negative relationships (**Table 1**).

Processing of soil fertility, as funded by the FNGLA, is mostly complete. We have organic matter (OM) and pH quantified for all study sites but are still awaiting total C and total N results for last field season's yards (60 of the total N=90). This delay is due to equipment issues in the lab processing our samples. We recently received notice that these final analyses are in process. Distribution of soil OM and its relationship with the maintenance intensity index (no evidence of relationship) is shown in **Figure 5**.

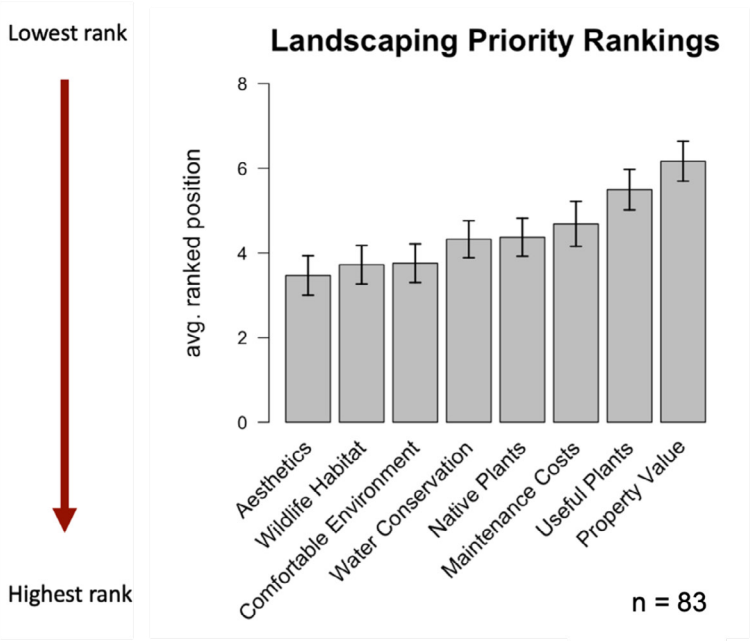


Figure 3. Results of ranking exercise performed by study participants of priorities when making landscaping decisions about their yards. Chart displays average ranked position, so lower values equate to higher overall prioritization (i.e., most to least important ordered left to right).

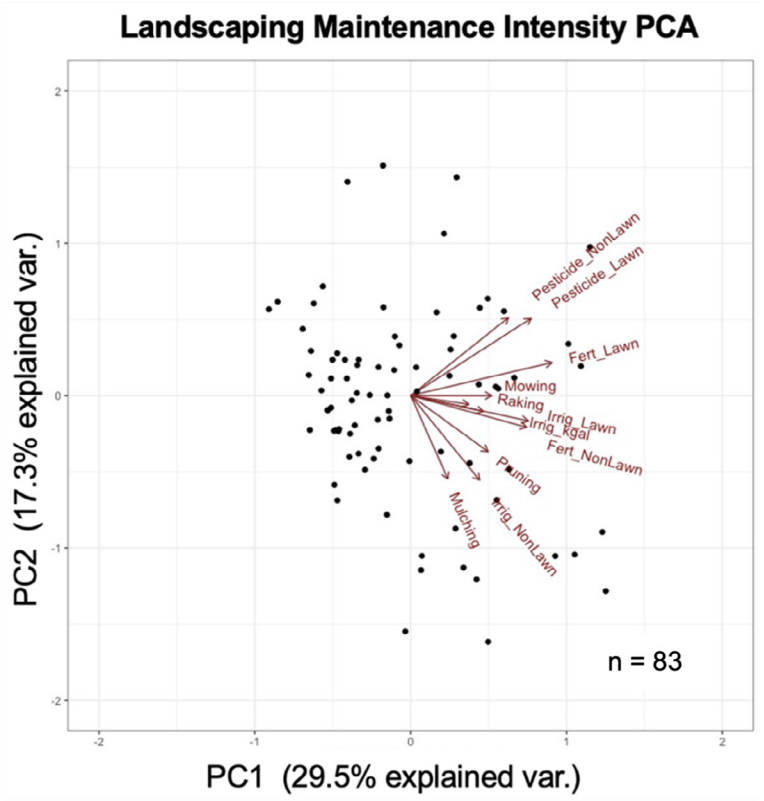


Figure 4. Principle component analysis of homeowner reported maintenance practices and outdoor water use. The main axis (PC1) is characterized by use of irrigation, fertilizer, and pesticides explaining approximately 30% of the variation in maintenance intensity.

Table 1. Summarized relationships between maintenance intensity index (PC1) described in Figure 4 and homeowner landscaping priorities depicted in Figure 3. Also included is the relationship with a preliminary assessment of soil fertility (shaded orange). Quantification of soil fertility is currently limited to soil OM, but will be refined when we receive results of total C and total N.

Ecosystem Service	Relation to Landscaping Maintenance Intensity
Aesthetics	→
Wildlife Habitat	↘
Comfort	↘
H_20 Conservation	↘
Native Plants	↘
Soil Fertility (OM)	→

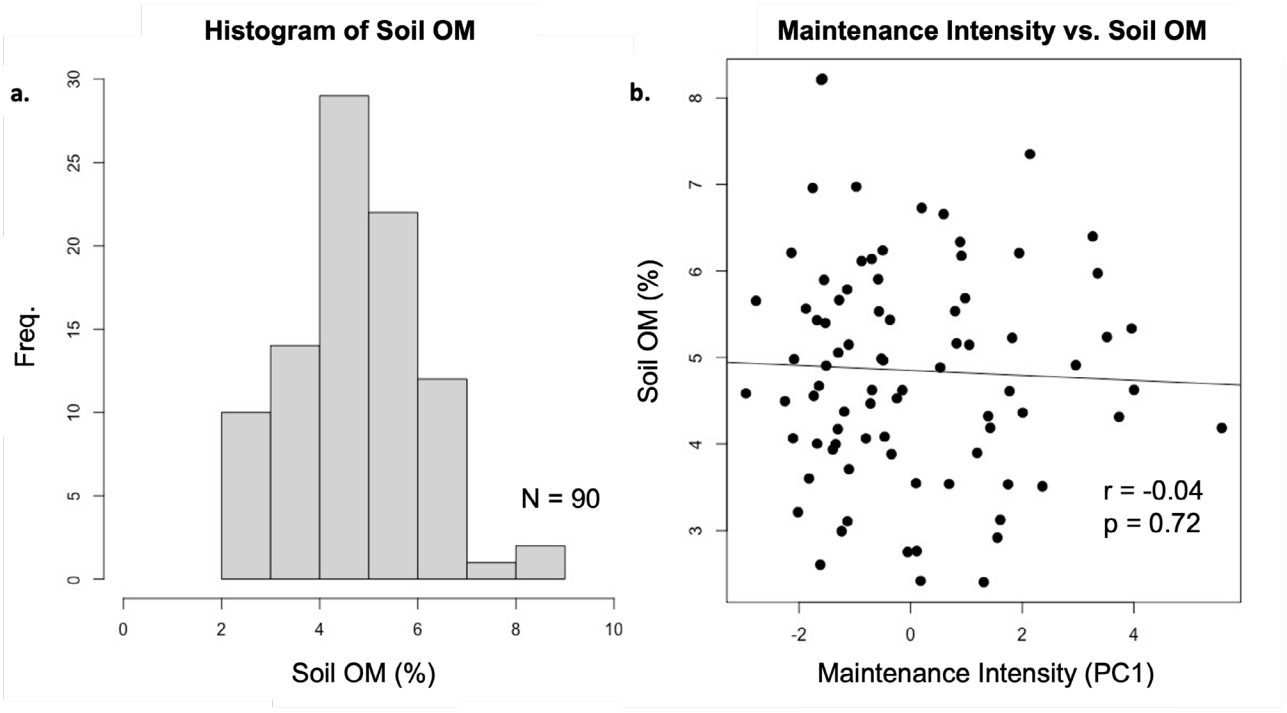


Figure 5. Distribution of soil OM across 90 Gainesville, FL yards (a) and regression analysis of soil OM as a function of maintenance intensity (b).

CONCLUSION AND NEXT STEPS

Initial results indicate evidence that landscaping maintenance intensity, characterized by irrigation, fertilizer, and pesticide use, is not achieving and even counterproductive to the desired landscaping outcomes of homeowners. This gap between what homeowners want and have implies marketing and educational opportunities for the landscaping industry to meet this demand and enhance quality of life for Floridians (FNGLA research priority #5) through providing desired ecosystem services. Insight from these relationships will improve with incorporation of more detailed soil fertility data once processing of total C and total N is complete. Moving forward, this complex and interdisciplinary dataset will be used to inform predictive models that relate ecosystem services to landscaping costs, management inputs/intensity, plant diversity, and vegetation structure. This will enable landscaping industry professionals and other constituents to make informed cost-benefit predictions for different landscape designs that promote a more sustainable future for Florida and its residents.

IMPROVE PEST MANAGEMENT PRACTICES AND STRATEGIES

This priority area is defined as:

FNGLA supports research to develop new biological and chemical pest management tools that are effective and environmentally safe.

FNGLA supported six projects under this priority area, and their summaries are on pages 12-43.

Assessment of insecticides for control of Haplaxius crudus, the vector of lethal bronzing

PI: **Brian Bahder, Entomology and Nematology, Ft. Lauderdale REC**

Co-PI: **De-Fen Mou, Entomology and Nematology, Ft. Lauderdale REC**

ABSTRACT

Lethal bronzing (LB) is a fatal phytoplasma infection of a variety of ornamental palms in Florida and is a significant threat to the green industries. Current management has emphasized rapid removal of infected palms and preventative inoculations of the antibiotic oxytetracycline (OTC) but little has been done in terms of management of the insect vector, *Haplaxius crudus*. In this study, the efficacy of three different formulations of imidacloprid were evaluated for their potential to kill adults of *H. crudus*. It was determined that a broadcast spray and soil drench were highly effective at killing *H. crudus* while the granular formulation showed no difference relative to the control palms. These findings are valuable in that they demonstrate the efficacy of imidacloprid against *H. crudus* but also show potential in reducing disease spread.

METHODOLOGY

Objective 1: Evaluate efficacy of 75WSP as soil drench against *H. crudus.*
Objective 2: Evaluate efficacy of 2F as broadcast spray against *H. crudus.*
Objective 3: Evaluate efficacy of Merit granular treatment against *H. crudus.*

Three different treatments were established on seedlings of *Phoenix sylvestris* (**Figure 1**) using imidachloprid based on formulation. Products used were Merit granular at 0.2g/0.02m2 per pot, 2F applied at turf rate as broadcast spray (0.6 oz/1000ft2), and 75WSP (17ml in 1 gallon of water, 250 ml applied per pot as a soil drench). Each formulation was replicated 10 times. 10 control pots with no insecticides were used for comparison and to assess natural mortality from bring wild caught individuals from the field. All plants were treated 2 weeks prior to addition of *H. crudus* to allow for insecticides to become systemic in plants. 20 adults were used per plant. Mortality was recorded hourly for first 6 hours then daily until all adults on control plants were dead.

RESULTS

Significant mortality in adults of *H. crudus* was observed within the first 24 hours for 75WSP and 2F treatments with 55% and 35% mortality within 6 hours, respectively, compared to 20% and 13% mortality for the control and merit treatment, respectively (**Figure 2**). At 24 hours post exposure, approximately 92% and 86% were dead for the 75WSP and 2F treatments, respectively, whereas there was about 42% and 34% mortality for control and merit treatment (**Figure 2**). For the 75WSP treatment, 100% mortality was attained at 4 days, 6 days for the 2F treatment and 17 days for the control and merit treatment (**Figure 2**).

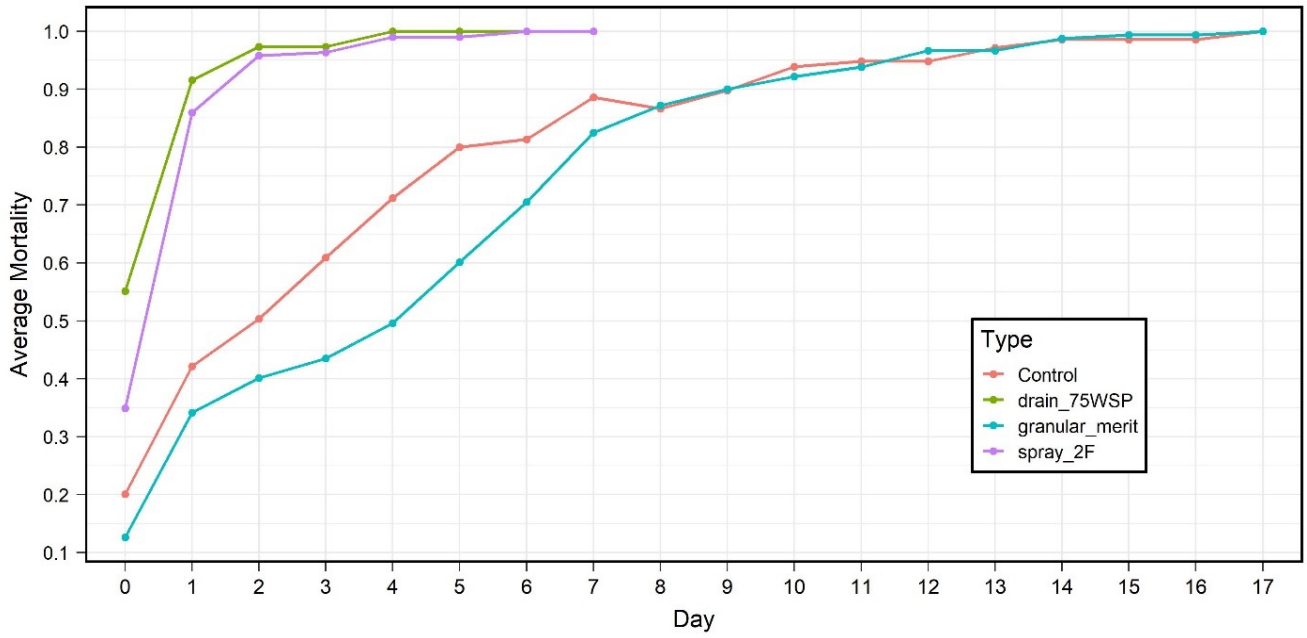


Figure 2. Mortality data for *H. crudus* for all three formulations of imidacloprid on palm seedlings of *Phoenix sylvestris*

CONCLUSIONS

These data indicate that imidacloprid is highly effective at killing *H. crudus* when applied to palm seedlings as a spray and soil drench but not effective in granular formulation. While these insecticides are registered for use on turf grass, these data are still highly relevant because the immature stages of *H. crudus* are strict grass feeders and persist in the thatch layer of a wide variety of grasses and sedges in Florida. The application of these insecticides on turf grass to control a variety of turf pests and will also target nymphs of *H. crudus*. In addition, if insecticides are applied near palms, the uptake of these insecticides in palms will also grant a degree of protection from infection by LB. It has been determined that optimal acquisition time for LB is 5 days and since the 75WSP killed 100% of *H. crudus* at 4 days, it indicates that palms with imidacloprid in the field will likely result in the death of *H. crudus* prior to it acquiring or transmitting any phytoplasma. Future studies need to assess injection technologies and assess uptake of the above formulations in other palm species to verify efficacy is consistent among different palm species important for the green industries of Florida.

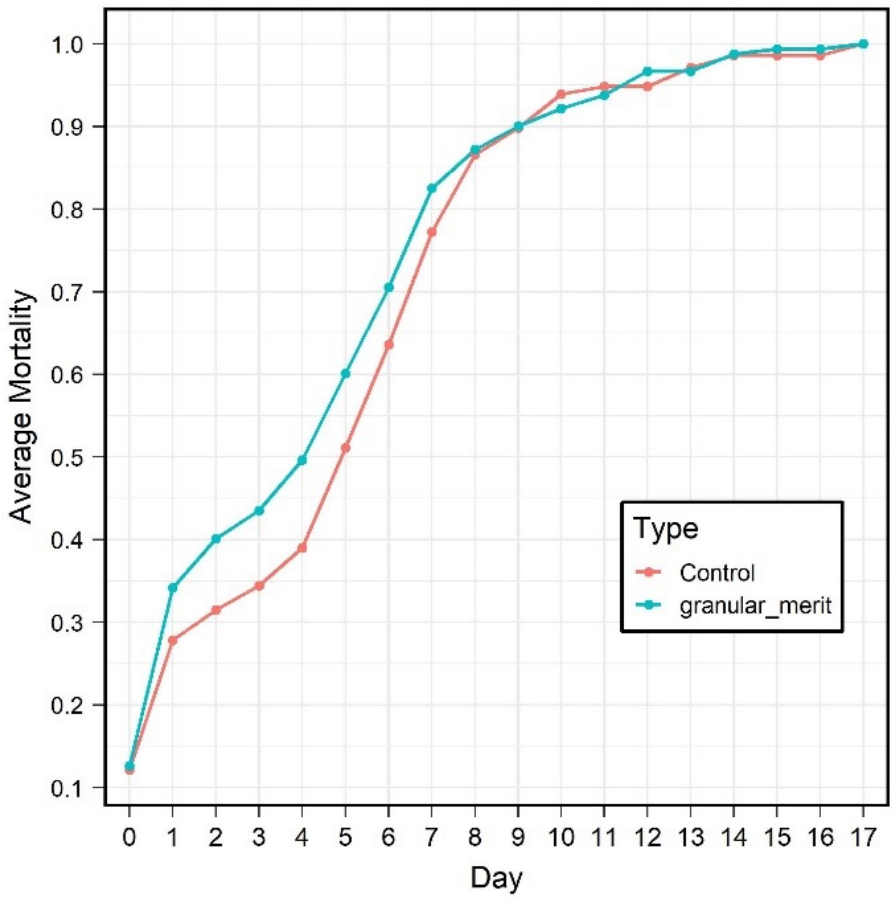


Figure 3. Mortality data for *H. crudus* for granular treatment versus control.

Detecting overlap of pathogen presence and trunk rot in palms

PI: **Braham Dhillon, Plant Pathology, Ft. Lauderdale REC**

ABSTRACT

Ganoderma butt rot of palms is a lethal disease caused by the wood-decaying white-rot fungus *Ganoderma zonatum*. This disease is a major concern for the landscape industry across palm-growing regions in the US as all palm species are considered susceptible. Decay caused by *Ganoderma* compromises the structural integrity of the trunk and increases the likelihood of palm failure threatening property damage and human safety. However, very little is known about the fungal movement and spread in the trunk and its correlation to trunk rot. Using the PCR-based species-specific assay we have developed for *G. zonatum*, it was determined that the fungus is present in diseased palms up to 3 feet above the soil line and a positive signal for *G. zonatum* could be detected at least five months before a conk was visible. The visual representation of trunk decay using sonic tomogram correlated to the actual rot in the trunk.

OBJECTIVES

1. Measure colonization zone of the fungus, *G. zonatum*, using species-specific molecular assay

2. Assess extent of rot damage inside the trunk using sonic tomography

Wood shavings were collected and used to detect the presence of *Ganoderma zonatum* in healthy and diseased palms. Briefly, the palm trunk circumference was marked into eight equal sections at 1-foot increments from the soil line up to a height of 3 feet. Wood shavings (at least 100 mg) were collected from each grid square using a 5/8" drill bit, resulting in 24 samples per palm every month. Samples were collected on a monthly interval from six foxtail palms over a period of six months. Additionally, one triangle palm and two foxtail palms were sampled once during the study. DNA was extracted from each sample and used for PCR-based detection of *G. zonatum* presence in the palm trunks. To determine rot inside the palm, acoustic tomograms were recorded from each palm close to soil line using the PiCUS^3 sonic tomograph (Argus Electronic GmbH).

RESULTS

A *Ganoderma zonatum* species-specific detection assay coupled with sonic tomography was used to screen healthy and diseased foxtail palms over a period of six months. The health status of the six foxtail palms (FTP1 – FTP6) initially selected for the study was determined by visual assessment of palm canopy and trunk regions. Two foxtail palms (FTP1 and FTP3), had extensive visual symptoms of Ganoderma butt rot, including reduced canopy, unopened fronds and spear leaf, and presence of conks. The remaining four foxtail palms (FTP2, FTP4, FTP5, and FTP6) appeared healthy based on green fronds, and full canopy. The health status of these palms was monitored visually throughout the sampling period.

Presence of *Ganoderma* in the palm trunk was detected using the PCR assay well before extensive damage due to rot could be visualized by the sonic tomography. Every sample collection site on the trunk was unique which contributed to the stochastic variation in fungus detection over the sampling time frame (**Figure 1**). The sites for monitoring trunk decay using sonic tomography were established at the start of the project and provided useful data for a period of six months.

The diseased palms, FTP1 and FTP3, showed a strong agreement between presence of *G. zonatum* and extensive trunk decay (**Figure 1**). In May 2022, foxtail palm FTP1 lost its canopy and died but stump remained intact and upright, whereas foxtail palm FTP3 had three unopened fronds. Sonic tomogram for FTP1 and FTP3 showed widespread trunk damage due to rot manifested by *G. zonatum* growth in the trunk (**Figure 1**).

Amongst the healthy foxtail palms, two (FTP4, FTP5) lacked *Ganoderma* conks, whereas the other two foxtail palms (FTP2, FTP6) had a conk present on the aerial roots. *Ganoderma zonatum* was undetectable in three foxtail palms, FTP2, FTP4 and FTP6, at the beginning of the study, while foxtail palm FTP5 was positive for *G. zonatum* presence.

The sonic tomogram for foxtail palms FTP2 and FTP6 matched the output from the PCR assay, whereas the tomograms from foxtail palms FTP4 and FTP5 were inversely related. Foxtail palm FTP5 showed extensive fungal growth but lacked detectable decay in the tomogram, while for foxtail palm FTP4, tomogram exhibited decay in the trunk core with it being positive for *G. zonatum* only in the last month of sampling.

Besides the routine monthly sampling of foxtail palms (FTP1 – FTP6), three additional palms, including two foxtail palms (FTP7 and FTP8) and one triangle palm (TAP1), were also evaluated during this study. The foxtail palm FTP7 was sampled close to soil line after two fronds were seen to rapidly desiccate within a period of two weeks. The PCR assay was positive for *G. zonatum,* and the sonic tomogram showed signs of decay in the trunk core (**Figure 2A**). Presence of *G. zonatum* in the trunk was evident from the appearance of fruiting body initials in July 2022 (**Figure 2A**).

Sampling of triangle palm TAP1 exhibiting typical symptoms of Ganoderma butt rot confirmed the presence of *G. zonatum* up to a height of 6 ft. in the palm trunk. Sonic tomogram showed extensive damage resulting from rot in the trunk. The loss of structural integrity was revealed by wind breakage and falling of the palm trunk to the ground one-week after sampling.

The foxtail palm FTP8 was suspected to be diseased as it did not respond to fertilizer applications over a period of two years. At the time of observation, the crown appeared yellowish green with an unopened spear leaf but there were no desiccating fronds (**Figure 2B**). No conks were visible on the trunk and the PCR assay did not detect *G. zonatum* in the trunk. The sonic tomogram revealed a healthy trunk without any rot (**Figure 2B**), which corresponded to the visual observation of the trunk cross-section after it was cut down (**Figure 2B**).

CONCLUSIONS

The *Ganoderma zonatum* specific assay was found suitable for detection of *G. zonatum* in palm trunk tissue before signs of decay could be detected either by visual symptoms i.e., desiccating fronds, or by using sonic tomography. Similarly, non-invasive visualization of trunk decay using sonic tomography was an accurate predictor of rot damage in the trunk. The entry and growth of *Ganoderma* in the palm trunk tissue is highly variable making detection very challenging. In order to increase the success of detection, we recommend sampling from two sites diagonally opposite on the trunk.

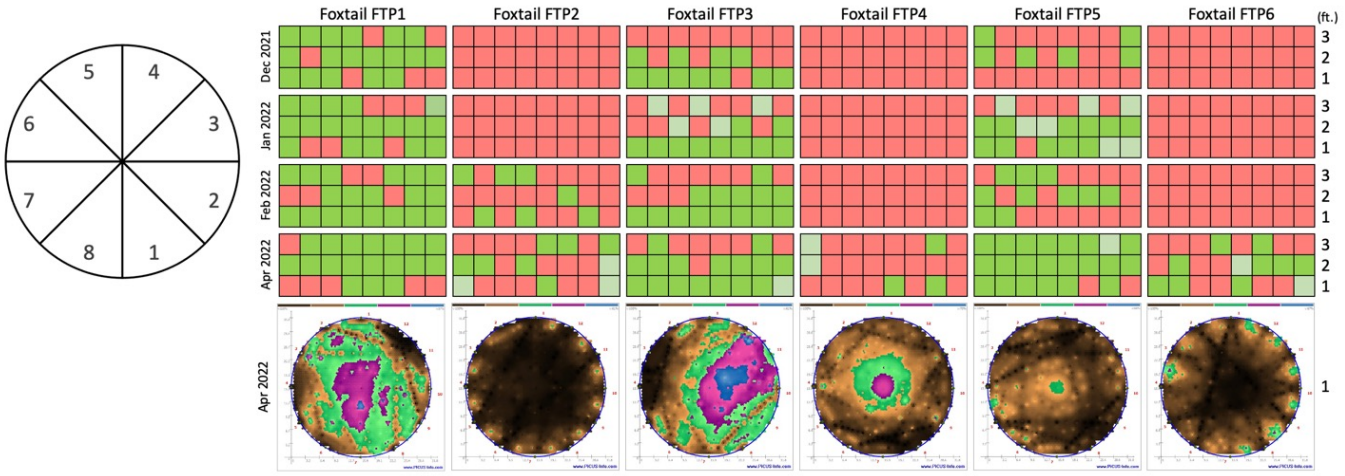


Figure 1. Ganoderma zonatum detection and sonic tomography of healthy and diseased foxtail palms.

The palm trunk was marked into eight equal sections, as shown in the pie chart on the left. Wood shavings were collected from each section at three heights, 1, 2, and 3 feet, from the soil line. A total of 144 samples were obtained from six foxtail palms (FTP1 – FTP6) in one month and used for DNA extraction. The presence and absence of *G. zonatum* in the trunk tissue determined by the species-specific PCR assay is represented by green and salmon color squares, respectively. The colors in the sonic tomogram (bottom row), starting from brown, tan, green, purple, and blue, signify an increasing loss of palm trunk integrity due to rot. Assessment of trunk decay from April 2022 is shown here. Sample collection and sonic tomography was conducted over a period of six months. The numbers on the right denote the height (in feet) from the soil line at which wood shavings were collected.

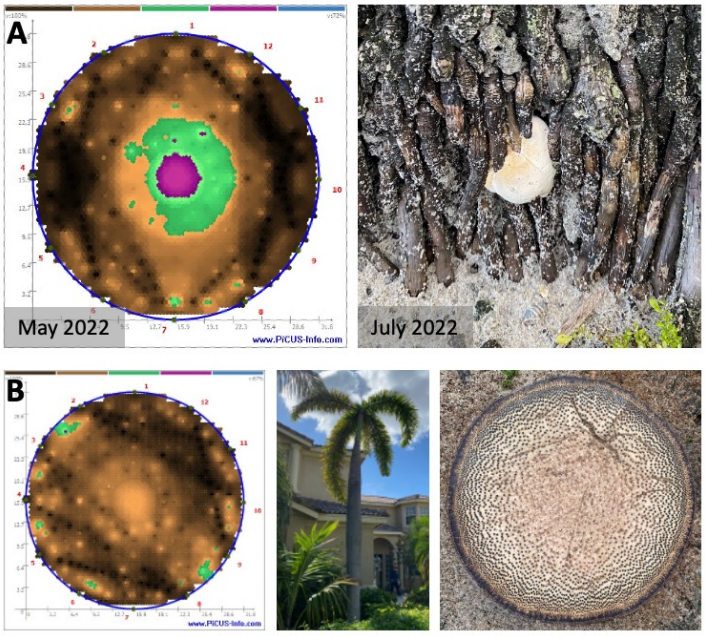


Figure 2. Assessment of rot in unhealthy foxtail palms.

A) Decay in the trunk core was documented by sonic tomography in foxtail palm FTP7 that had two desiccating fronds, and presence of *G. zonatum* was confirmed the appearance of fruiting body initials. B) Foxtail palm FTP8 suspected of decay in trunk was found to be free of rot by sonic tomography and trunk cross-section.

Integrating pest and pollinator management strategies for ornamental plant production

PI: **Adam G. Dale, Entomology & Nematology**

Co-PIs: **Jaret Daniels, Florida Museum of Natural History**
Bernadette Mach, Entomology & Nematology

ABSTRACT

Demand for wildlife-friendly plants, especially plants that support pollinators, has never been higher. This is great because producing and planting wildlife-friendly plants helps mitigate biodiversity loss, which is an increasing global concern. This demand also presents an incredible business opportunity for Florida's green industry. Unfortunately, many plants that support pollinators are also attacked by damaging insect pests. Insect pests reduce plant health and marketability, which often requires chemical intervention to produce a saleable product. Plant pests may also directly reduce the conservation value of plants by reducing plant quality and competing with pollinators. Most pollinator toxicological data focus on bees. Lepidopteran pollinators (i.e., butterflies and moths) differ in susceptibility to insecticides compared to bees and have different routes of exposure (e.g., larval leaf-feeding versus nectar consumption). Consequently, products compatible with bee conservation pose an unknown risk to lepidopteran pollinators, yet both taxa are pervasive in agroecosystems and urbanized landscapes. We used the milkweed-monarch-oleander aphid system as a model to begin developing integrated pest and pollinator management (IPPM) strategies for ornamental plant production. Our results demonstrate that all commercially used systemic insecticides targeting aphid pests have toxic effects on monarch caterpillars at some timepoint after application. However, these effects and the longevity of effects vary significantly between insecticides. More specifically, we found that acute mortality from exposure to treated milkweed for 48 hours occurs with most evaluated insecticides, but only within the first week after application. More importantly, we find that all insecticides have toxic effects from chronic exposure, which leads to monarch death at the pupal stage. Some products have this effect up to 28 days after treatment. This is notable, because it suggests that most insecticide mortality to monarchs occurs out of site and thus out of mind since monarchs typically wander away from their host plant to pupate. Results will be used to develop a certified wildlife-friendly plant production protocol that will increase the positive environmental and ecological impact of ornamental plant production.

OBJECTIVES

1. Determine the effects of aphid infestation on monarch conservation outcomes.
2. Determine the acute and chronic exposure toxicity of commonly used and proposed alternative insecticides to monarch butterfly larvae.
3. Evaluate the effectiveness of commonly used and proposed alternative insecticides to control oleander aphid infestations on tropical milkweed.

METHODS

The Study System

Tropical milkweed is a nonnative, herbaceous perennial plant that is widely available for purchase across the eastern and southern United States and is the most popular milkweed species for landscape and garden use. The monarch butterfly (*Danaus plexippus*, L.) is a widely recognized charismatic North American butterfly that engages in a yearly migration from the eastern United States and Canada to overwintering grounds in Mexico. Monarch larvae are dietary specialists that feed exclusively on milkweed (family Apocynaceae, subfamily Asclepiadoideae). Monarch populations have declined by over 80% in recent decades (Brower et al. 2011, Thogmartin et al. 2017, USFWS 2020a, b), which has spurred widespread conservation efforts, primarily in the form of providing larval host plants. The oleander aphid (*Aphis nerii*, Fonscolombe, 1841) is an important sap-feeding insect pest of milkweed and other related ornamental plants in tropical to warm temperate regions of the world. Milkweed infested with oleander aphids become chlorotic, drops leaves, and are often covered with black sooty mold, ultimately resulting in an unsaleable plant. Thus, insecticides are commonly used during production to prevent plant damage and loss. Since many insecticides are toxic to Lepidoptera, milkweed treated for aphid infestations may inadvertently control monarch larvae or other beneficial caterpillar species during plant production and after sale. Consequently, plants produced and planted for wildlife conservation purposes may have the opposite effect. This combination of factors makes the monarch butterfly-milkweed-oleander aphid system ideal for developing IPPM strategies compatible with lepidopteran larvae conservation.

Objective 1 approach

An important initial question is: Do oleander aphids interfere with monarch conservation? We hypothesizedthat high-density aphid infestations would have negative effects on monarch oviposition and larval success by reducing plant quality and upregulating plant defenses. We measured the number of eggs deposited on milkweed plants by placing one adult male and one adult female in a mesh cage with one aphid-free and one aphid-infested tropical milkweed plant. Monarchs were allowed to oviposit freely and all eggs on each plant were collected and counted seven days later.

For the larval feeding study, we assigned third instar monarch larvae to three aphid density treatments: aphid-free milkweed leaves, aphid-infested milkweed leaves with sooty mold, aphids, and honeydew (i.e., "dirty" leaves), and aphid-infested milkweed leaves that had been cleaned with water to remove sooty mold, aphids, and honeydew (i.e., "cleaned" leaves). Each larva was placed in a Petri dish with one leaf and stored at 28 °C and 75% relative humidity with a 12:12 L:D diurnal cycle for seven days. A single new milkweed leaf was added to each Petri dish when > 50% of the old leaf was consumed or every 48 h, whichever came first. Total leaf area consumed (cm^2) per larva was calculated. Final instar attained and larval weight were also measured after seven days.

Objective 2 approach

Three industry standard insecticides (imidacloprid, spirotetramat, and insecticidal soap) were selected based on a 2017 Florida nursery grower survey. We also selected three proposed alternatives (pymetrozine, acetamiprid, and flupyridifurone) that are labeled for aphid control on ornamental plants and suspected to provide reduced risks to lepidopteran insects. Each insecticide was applied at the labeled aphid control rate. Selected insecticides and their properties are detailed in **Table 1**.

Our first question was – Do any of these insecticides have toxic effects on monarch caterpillars due to acute exposure, or within 48 hours of feeding on treated plant tissue, and how does this change over time after treatment application? For this acute toxicity experiment, six milkweed plants were randomly assigned to each treatment. Plants were treated with the appropriate insecticide via foliar sprays at labeled rates for aphid control. To assess acute oral toxicity associated with each insecticide, we exposed third-instar monarch larvae to treated tropical milkweed leaves for 48 h. Leaves were harvested 24 h, two weeks, and four weeks after insecticide treatment and placed individually in a Petri dish with a single third-instar monarch larva and a piece of damp filter paper. Petri dishes were held at 28 °C and 75% relative humidity with a 12:12 light cycle. Larval mortality (%) and leaf area consumed were measured at 48 h in the Petri dish.

Our second question was – Do any of these insecticides have toxic effects on monarch caterpillars due to chronic exposure, or when caterpillars are allowed to feed on treated plants until they pupate and complete development, and how does this change over time after treatment application? For this chronic toxicity experiment, eighteen plants were randomly assigned to one of the six treatments and further subdivided into three cohorts of six plants that designate exposure at different time points after treatment (24 H, 2 weeks, 4 weeks). These time cohorts corresponded with times post-treatment at which monarch larvae were added to the plants and given the opportunity to feed and complete development. All plants were treated at the same time via foliar spray with the appropriate insecticide at labeled rates for aphid control. At 24 h, 2 weeks, and 4 weeks post-treatment, a single third-instar monarch larva was placed on each plant in the appropriate treatment × time cohort. Monarch mortality was assessed until all larvae either perished or reached adulthood.

Table 1. Selected insecticides and their properties.

Insecticide	IRAC mode of action	Application Rate (g or ml pesticide per 379 L H_2O)	Active Ingredient (g or ml per 379 L application)	Labeled for caterpillars	Honey bee contact toxicity rating	Honey bee oral toxicity rating
Pymetrozine[1]	9-B	142 g	71 g	No	Practically nontoxic	Practically nontoxic
Spirotetramat[1,2]	23	100 ml	24 g	No	Practically nontoxic	Practically nontoxic
Acetamiprid[1]	4-A	118 ml	11 g	Yes	Practically nontoxic	Moderate
Flupyradifurone[1]	4-D	298 ml	60 g	No	Practically nontoxic	Moderate
Imidacloprid[2]	4-A	50 ml	12 g	Yes	High	High
Insecticidal soap[2]	-	7393 ml	3661 g	Yes	n/a	n/a

[1]Reduced Risk, US EPA
[2]Industry standard

Objective 3 approach

Since insecticide applications are made to milkweed to suppress oleander aphid infestations, we also evaluated the effect of each insecticide on aphid density per plant during the chronic toxicity experiment. The severity of aphid infestation was rated once weekly beginning at the time of insecticide application and continuing for the duration of the chronic toxicity experiment. Aphid densities were averaged across all terminal growth points per plant and ranked on an ordinal scale as 0 (no live aphids), 1 (< 50 aphids per terminal growth point), 2 (approximately 50-150 aphids per terminal growth point), or 3 (>150 aphids per terminal growth point). Average aphid ratings above 1 indicate plants with infestations over the recommended treatment threshold of >50 aphids per terminal growth point.

RESULTS

Objective 1

We found that high-density oleander aphid infestations have substantial negative effects on monarch oviposition and larval health. High-density aphid infestations reduced oviposition, larval weight, and larval leaf consumption by nearly 50% (**Figures 1 & 2**). Taken together, the effects of high-density aphid infestations on monarchs have significant implications for conservation efforts in urbanized landscapes and emphasize the importance of controlling aphid infestations to preserve the conservation value of milkweed.

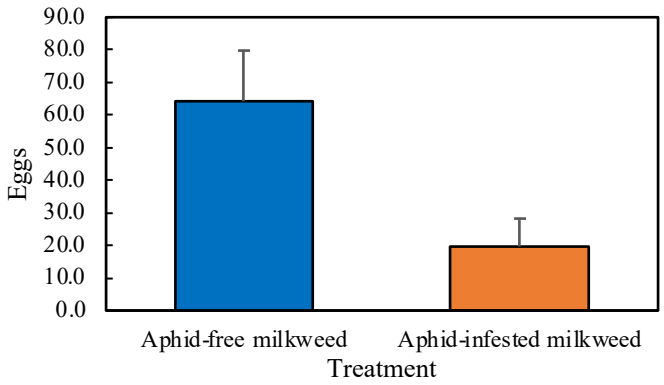


Figure 1. Mean (± SE) eggs laid by monarch butterflies on aphid-free versus aphid-infested milkweed. Different letters over bars indicates a significant difference (least-squares means, p<0.05).

Objective 2

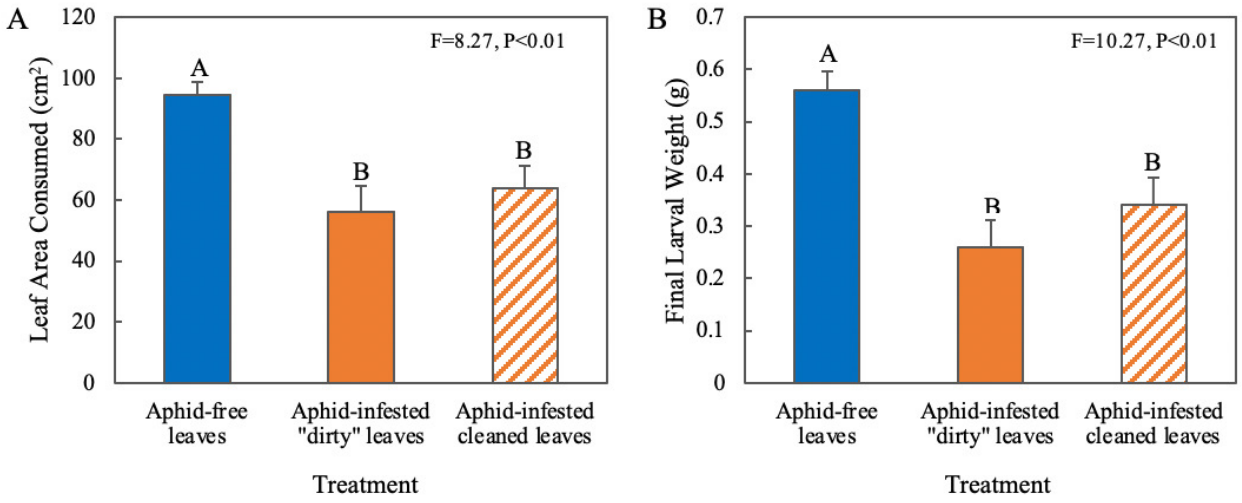


Figure 2. Mean (± SE) leaf area consumed (A) and final larval weight (B) of monarch larvae fed aphid-free milkweed leaves versus aphid-infested "dirty" leaves and aphid-infested cleaned leaves. Bars not topped by the same letter indicate significant differences (least-squares means, p<0.05).

Each of the six insecticides we evaluated resulted in either acute exposure mortality, chronic exposure mortality, or both to monarch larvae. Although spirotetramat and flupyridifurone are not labeled for caterpillar control, and pymetrozine is labeled as aphid-selective (Harrewijn & Kayser 1996), all products caused at least 40% monarch mortality at two or more of the chronic exposure test intervals. All three industry standard insecticides (imidacloprid, insecticidal soap, spirotetramat) caused at least 40% monarch mortality during two or more of the chronic exposure test intervals. Although we observed overall low mortality rates in the acute exposure trials, our chronic exposure results highlight an important and often cryptic consequence of non-target exposure to insecticides. Monarch mortality was higher when chronic exposure to treated milkweed plants was monitored for up to four weeks post insecticide application. Importantly, most of the chronic exposure mortality occurred as unsuccessful pupation, with larvae appearing to develop normally until fifth instar. This is important because it emphasizes the importance of tracking non-target impacts well past initial exposure and highlights that non-target effects of insecticides may be often out of sight and likely miss detection. Although we did not detect significant differences in mortality between treatments at 2 and 4 weeks, several insecticides caused 40% or greater mortality at both time points. These results demonstrate that insecticides commonly used during plant production pose significant risks to lepidopteran pollinators for periods well beyond application.

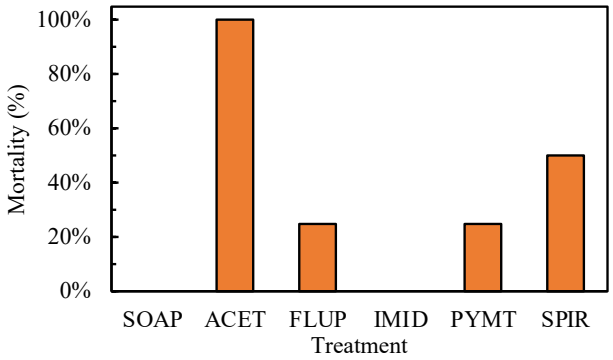


Figure 3. Acute exposure percent mortality for monarch larvae exposed to field-weathered insecticide residues for 48 h at 24 hours after insecticide application. No mortality was observed across all treatments at the 2 weeks and 4 weeks post treatment timepoints. Letters indicate significant differences between insecticides as determined by Kruskal-Wallis test with separation by post-hoc Dunn's test for multiple comparisons.

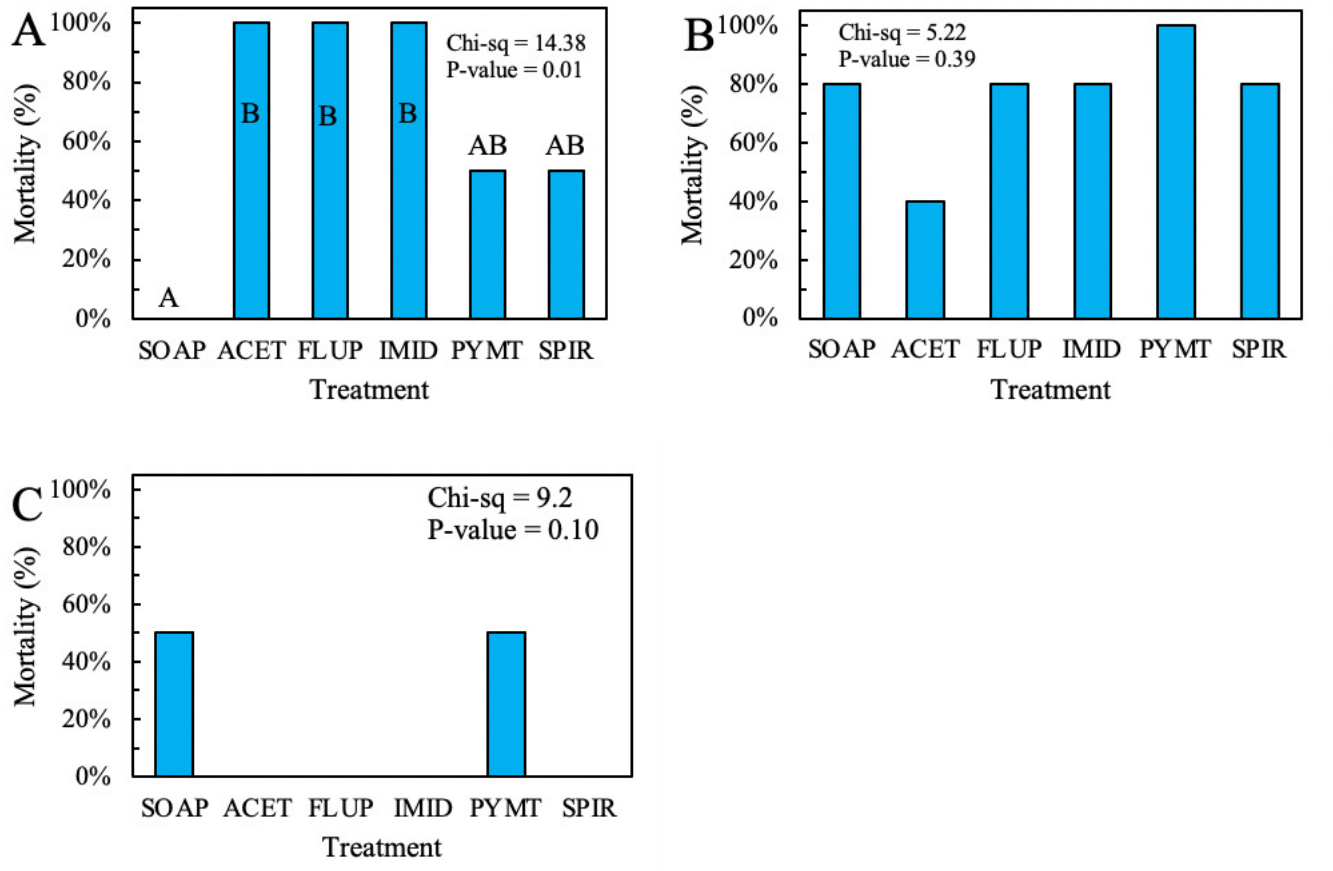


Figure 4. Chronic exposure percent mortality for monarch larvae exposed to field-weathered insecticide residues and provided the opportunity to complete development at A) 24 hours, B) 2 weeks, and C) 4 weeks after insecticide application. Letters on each bar indicate significant differences between treatments at each timepoint as determined by Kruskal-Wallis test with separation by Dunn's test for multiple comparisons.

Objective 3

Despite their commonplace use, none of the industry standard insecticides suppressed aphid densities below threshold levels for more than two weeks after one application. However, we two of our alternative products, flupyridifurone and acetamiprid, suppressed aphid densities below threshold levels for at least four weeks. Although acetamiprid is a neonicotinoid and flupyridifurone is neonicotinoid adjacent, each of these products are reduced risk insecticides and were among the best performers regarding non-target impacts to monarchs. These results suggest that nursery industry professionals should adjust their management programs to increase aphid control efficacy and reduce the required frequency of insecticide applications.

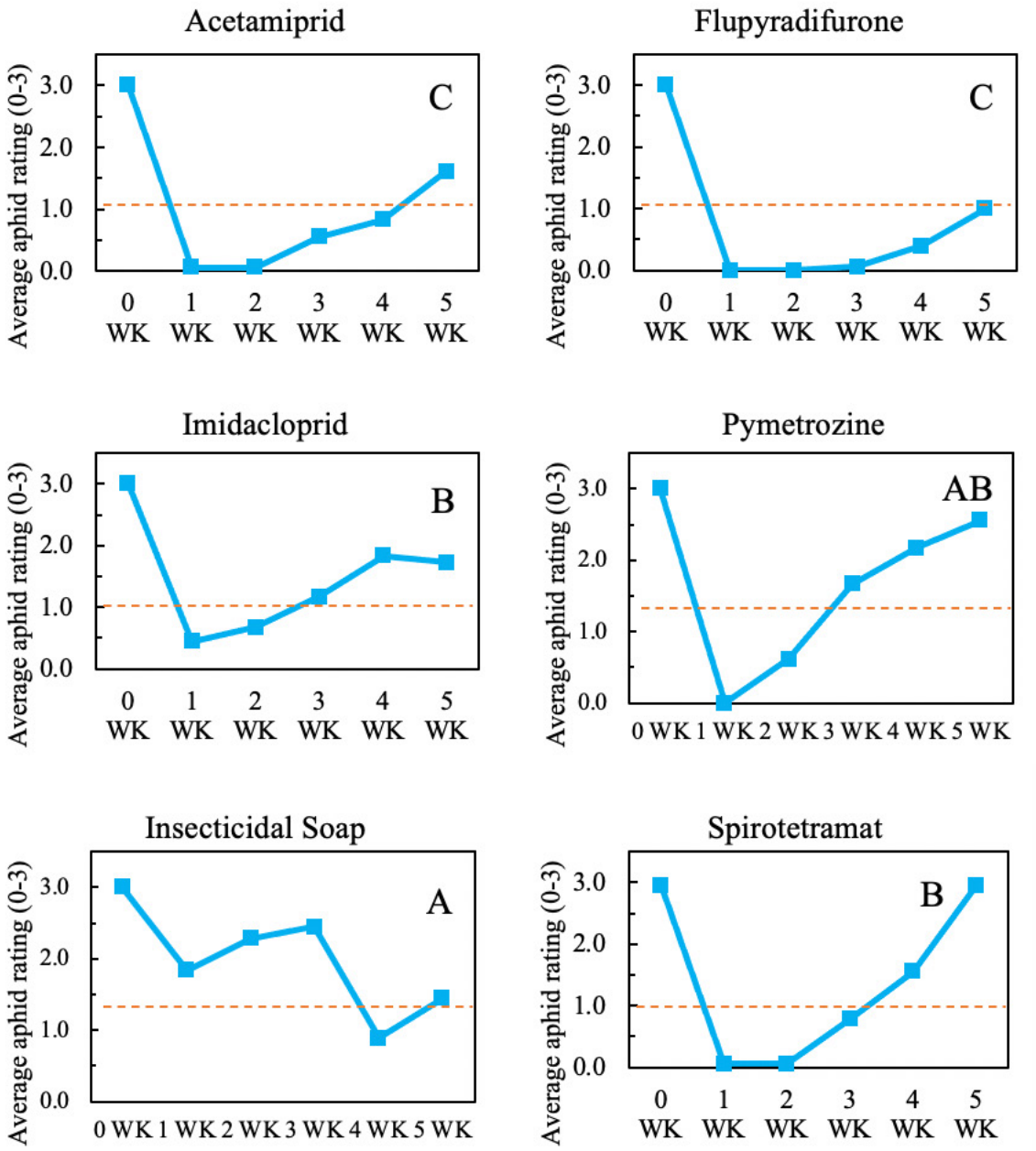


Figure 5. Average weekly aphid density rating (0-3) per treated milkweed plant during the chronic exposure trials. Treatment threshold (1) is indicated by the dashed line. Letters indicate significant differences between treatments over the duration of the experiment as determined by Kruskal-Wallis test (chi-sq = 80.076, p-value = 8.092e-16) with separation by Dunn's test for multiple comparisons.

CONCLUSIONS

Our results show that the effects of high-density aphid infestations on monarchs have significant implications for conservation efforts in urbanized landscapes. Pest-ridden milkweed provides less conservation value for monarchs as it recruits fewer monarch eggs, and those eggs will hatch into larvae with worse developmental outcomes, which in turn will result in adults with reduced fitness and migratory ability. This is especially relevant for milkweed plantings in urbanized landscapes where public conservation efforts are most prevalent, but where outbreaks of sap-feeding herbivores are also commonplace and difficult to manage. Unfortunately, insecticides used to control pests pose direct risks to monarchs. Furthermore, the duration of aphid control in these experiments was often shorter than the duration of adverse effects on monarch larvae, clearly demonstrating the conflict between aphid control and monarch larval conservation in ornamental plant production settings. Of the six insecticides tested during the chronic exposure trials, only two had a period where aphids were controlled below the treatment threshold and monarch larvae experienced no adverse effects. Additional research to develop IPPM strategies is urgently needed to ensure that milkweed and other lepidopteran host plants retain their conservation and monetary value during production.

"Place it and forget it" - Super absorbent medium for long-term weed suppression and plant-safe herbicide placement in nursery production.

PI: **Ramdas Kanissery, Horticultural Sciences, Southwest Florida REC**

ABSTRACT

Container production has several favorable factors for weed growth, making weed control challenging in potted plants. A prolonged weed suppression efficacy will be potentially achieved through pre-emergence herbicides; however, herbicide application in pots has to be attained through conventional spraying. These herbicide sprays can cause non-target effects through potential spray drifts, creating a non-conducive environment for nursery plants. Hence in this project, we explored the possibility of using a super-absorbent medium as a slow-release carrier for an effective and crop-safe placement of pre-emergence herbicide in potted plants. Observations from our experiment suggest that mixing herbicides with a super absorbent medium such as hydrogel could be a potential strategy for placing pre-emergence herbicides for suppressing weeds in container-grown crops and plants. However, additional research is needed to evaluate the crop safety of this strategy in plants with extreme susceptibility to herbicide-related injuries.

OBJECTIVES AND METHODS

The objective of this study was to evaluate the utility of super absorbent mediums such as 'hydrogel' for effective and plant-safe placement of pre-emergent herbicides in planter pots.

Justification

Weed management has become a pressing issue in container production as heavy fertilization, and constant irrigation creates a favorable environment for weed growth in containerized plants. Weeds compete with the plants for resources and potentially reduce the vigor and marketability of the container plants. There is heavy reliance on hand weeding, which is labor-intensive and, hence, not cost-effective. Recently, attention has been directed to using slow-release fertilizers in the nursery industry, whose benefits have been well documented. This concept has been applied to herbicide placement in containers by utilizing slow-release carriers or super absorbent mediums for pre-emergent herbicides.

When mixed with pre-emergent herbicide, this absorbent material will release only a part of the active ingredient in an immediately available form; the bulk of the herbicide is contained in an inert medium and is gradually released over time. Additionally, the herbicide retention by absorbent will potentially prevent the active ingredient from leaching into the root zone of the containerized plant (**Figure 1**). Beneficial effects of using pre-emergent herbicide in combination with a slow-release carrier medium in container production include a reduction in the amount of herbicide, a decrease in the risk of injury to the desired plants, cost savings by reducing the number of applications, and increased environmental safety. These super absorbent media is low-cost, ubiquitous, and typically biodegradable. Some of these materials are already used for hydro-mulching and as transplant amendments in nursery production. However, these materials' efficacy in absorbing and gradually releasing the active herbicide ingredients in plant containers remains understudied.

Methodology

Experiments were conducted in a greenhouse at the Southwest-Florida Research and Education Center in Immokalee, FL, in 2022. Planter pots (1.9 liters) were filled with a potting mixture (peat moss/pine bark/perlite) and planted with tomato (*Solanum lycopersicum L.*) and onion (*Allium cepa*) transplants. Each pot was also spiked with an equal number of seeds of broadleaf and grass weeds/plants (sun hemp, Bahiagrass, and ryegrass) to evaluate the weed suppression efficacy of the treatments. The super absorbent medium, also known as hydrogels or water retention granules (Stockosorb 660®), was thoroughly mixed with the pre-emergent herbicide solution containing indaziflam (e.g., trade name: Marengo) at a 20:1 ratio (i.e., 20 ml herbicide solution to 1 g of the absorbent medium). The indaziflam and absorbent medium mixture were placed in plant pots as a "top dress" **(Figure 2**). The treatments evaluated also included spray application of the herbicide solution, absorbent medium alone, and untreated control. A herbicide application rate within the labeled range for nursery production was used in the super absorbent medium mixture and herbicide sprays. Each treatment was replicated five times, and following treatments, pots were arranged in randomized complete block design on the greenhouse bench. The potted plants received regular overhead irrigation. At 4 weeks after treatment, aboveground biomass was collected by clipping the weeds and plants at the soil line and weighing them on a portable balance to assess the weed suppression efficacy and the growth vigor of potted plants.

Statistical analysis

All data were tabulated in Microsoft excel and tested using PROC GLM in SAS v9.4 (SAS Institute, Cary, NC). The data were tested for the assumptions of the linear model and log-transformed before subjecting to the analysis of variance (ANOVA) to test statistically significant differences between the means. The means were separated using Tukey's Honest Significant Difference (HSD) (α=0.05).

RESULTS

Applying pre-emergence herbicide mixed with a super absorbent medium reduced the weed pressure in the pots planted with onion and tomatoes (**Figure 3**). Although not statistically significant, compared to the untreated control, when herbicide was applied in combination with super absorbent medium, there was an 89% and 74% reduction in the biomass of weeds, respectively, in the pots planted with onions (**Figure 4**) and tomatoes (**Figure 5**). Additionally, the herbicide applied as a mixture with super absorbent medium outperformed the application of conventional sprays into the pots to control weeds in potted onion and tomato containers. Also, it is essential to note that the super absorbent medium alone in pots increased the weed pressure compared to untreated controls (**Figures 4 & 5**). This is because super absorbent mediums like hydrogels evaluated in this study can improve the soil water holding capacity and may have stimulated the weed germination and growth in the container.

The impact of test treatments on the growth and vigor of plants in the pots was also assessed. The aboveground biomass of onion and tomato plants is presented in **Table 1**. The herbicide applied in combination with super absorbent medium has lower herbicide phytotoxicity to the onion plant in the pot, as observed from its significantly higher biomass (18.1 ± 3.36 g) compared to the plants in the pots that received herbicide application as sprays (4.3 ± 1.78). However, the tomatoes were injured, and the biomass was considerably reduced from the herbicide + super absorbent mixture. Generally, tomatoes are easily susceptible to herbicide-related injuries, and a reduction in plant vigor and growth usually occurs when the plant is exposed to herbicides through direct contact, root uptake, etc. This observation suggests that the herbicide application strategy needs to be evaluated more for use in pots where sensitive crops such as tomatoes are planted.

CONCLUSIONS

Observations from this initial screening trial suggest that super absorbent medium such as hydrogels could be used as a carrier for placing pre-emergence herbicides for suppressing weeds in container-grown crops and plants. In containerized onions, weeds were suppressed effectively compared to control; moreover, the herbicide-related phytotoxicity was reduced as observed from the higher biomass compared to herbicide spray applications. Although enhanced weed suppression was attained in the potted tomatoes, plant growth was impacted by the herbicide applied in combination with super absorbent medium.

Additionally, the evaluated herbicide placement strategy in combination with a super absorbent medium facilitated a relatively effortless application of herbicides in the container compared to conventional herbicide spraying. This process also reduced the wastage of herbicide materials and labor. Moreover, the herbicide sprays also can potentially drift to surrounding areas in the nursery. However, such contaminations of the surrounding nursery infrastructure can be potentially minimized by adopting this herbicide placement strategy. Furthermore, additional research is needed to determine if this strategy can be utilized as a plant-safe method for herbicide placement in containerized herbicide-sensitive crops such as tomatoes and other solanaceous plants. Hence, there is a need to continue similar studies using other herbicide-active ingredients and a variety of containerized plants.

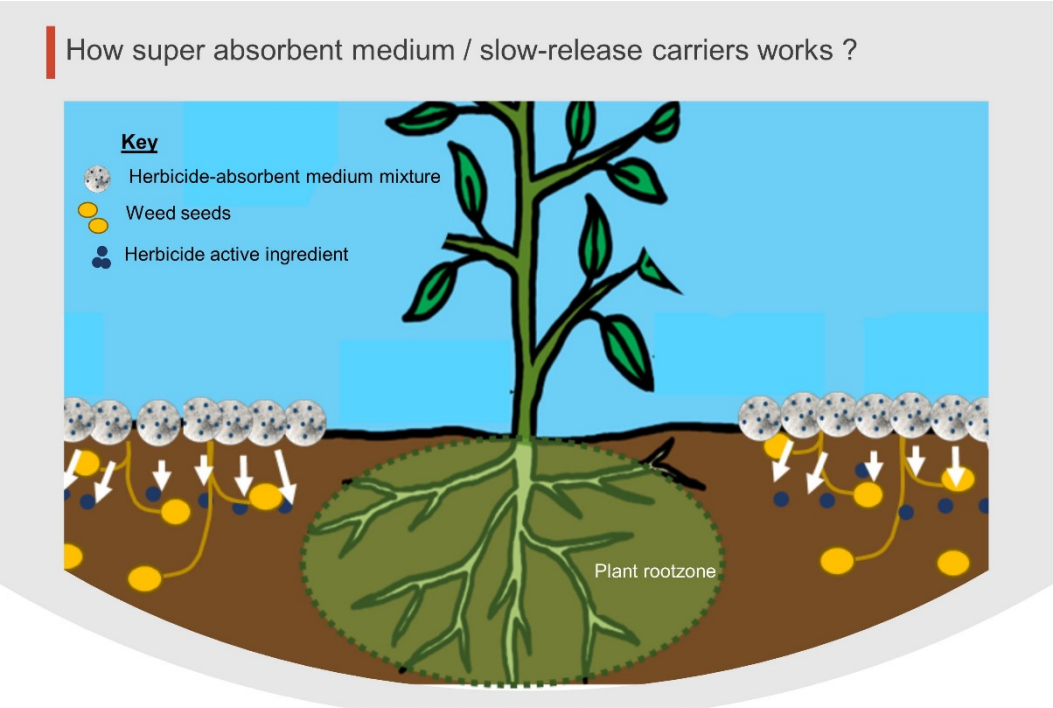


Figure 1. Schematic illustration showing the slow release of herbicide active ingredient from the herbicide-super absorbent medium mixture

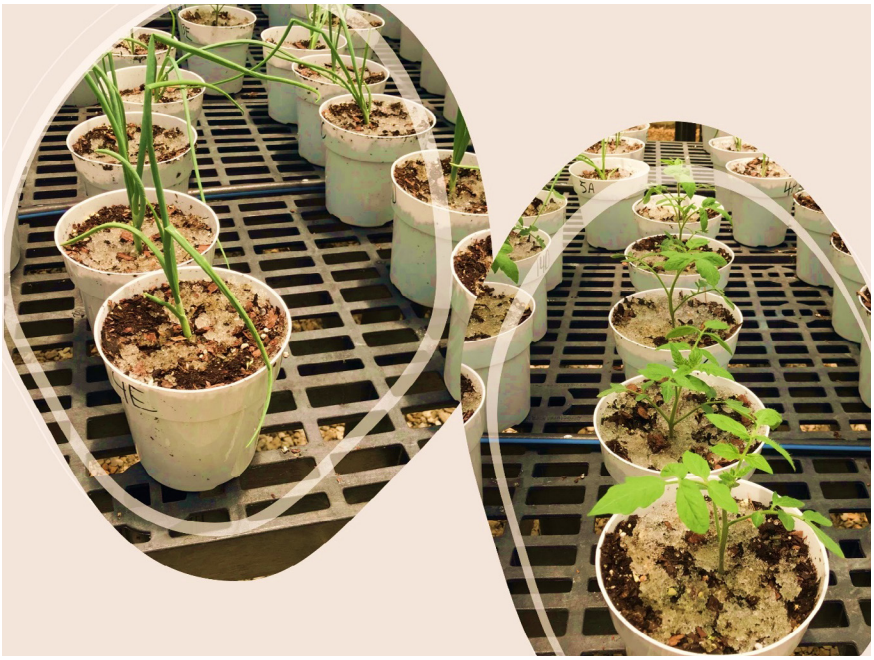


Figure 2. Pre-emergence herbicide (Indaziflam) + super absorbent medium mixture applied as 'top dress' in pots planted with onions (left) and tomatoes (right)

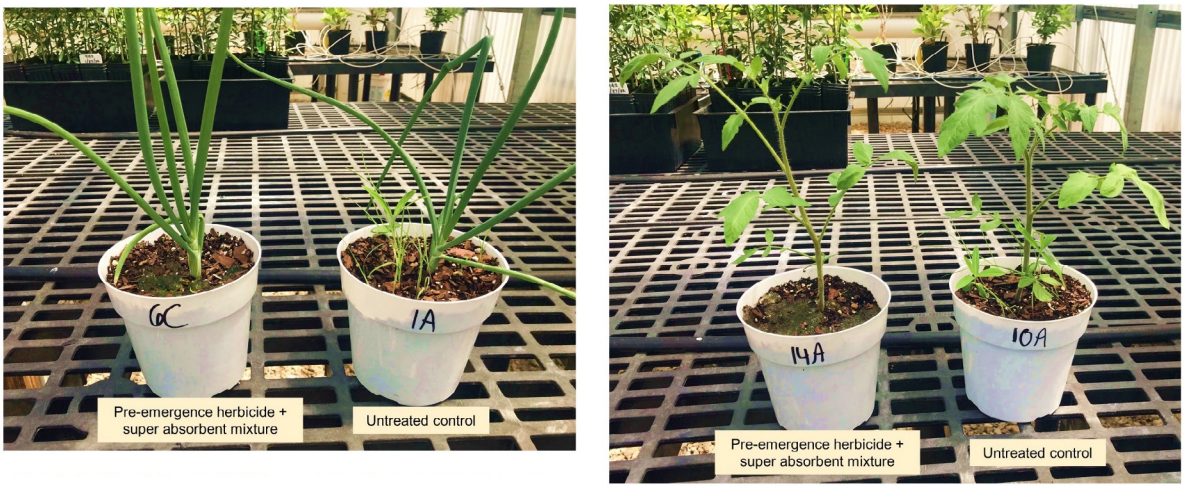


Figure 3. Weed suppression in potted onion (above) and tomato (below) from pre-emergence herbicide in combination with the super absorbent medium. Notice the weed growth in untreated control pots.

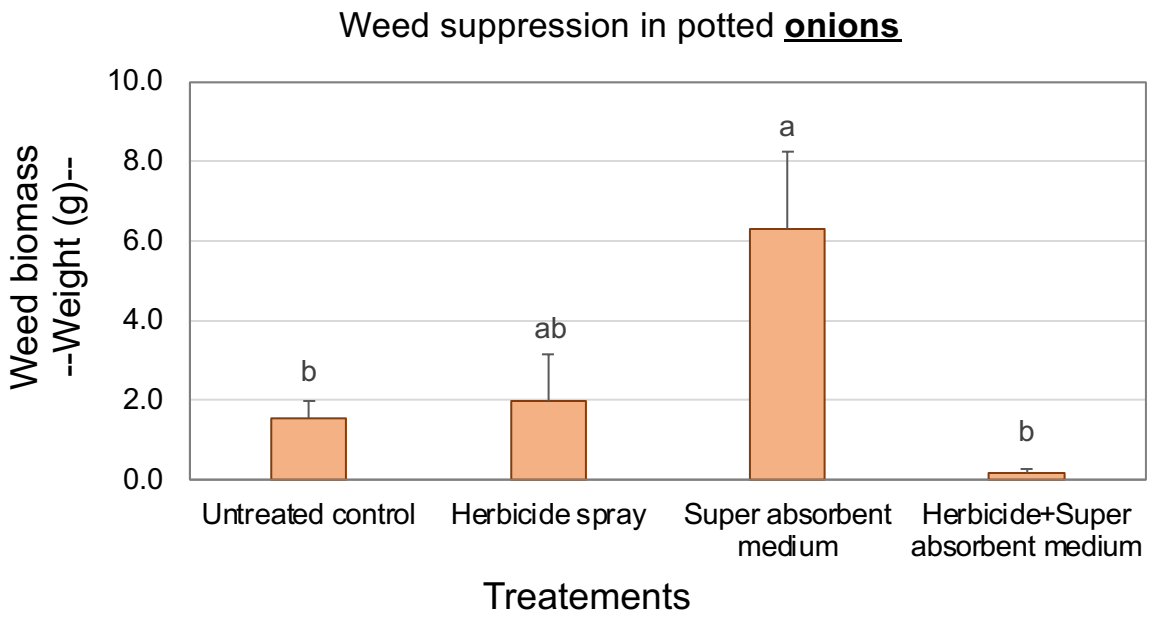


Figure 4. Effectiveness of treatments on suppressing weeds in pots planted with onions. Biomass of weeds measured from the treated pots 4 weeks after planting. Bars with the same letters do not significantly differ based on Tukey's Honest Significant Difference (HSD) (α=0.05). Error bars indicate the standard error of 5 observations.

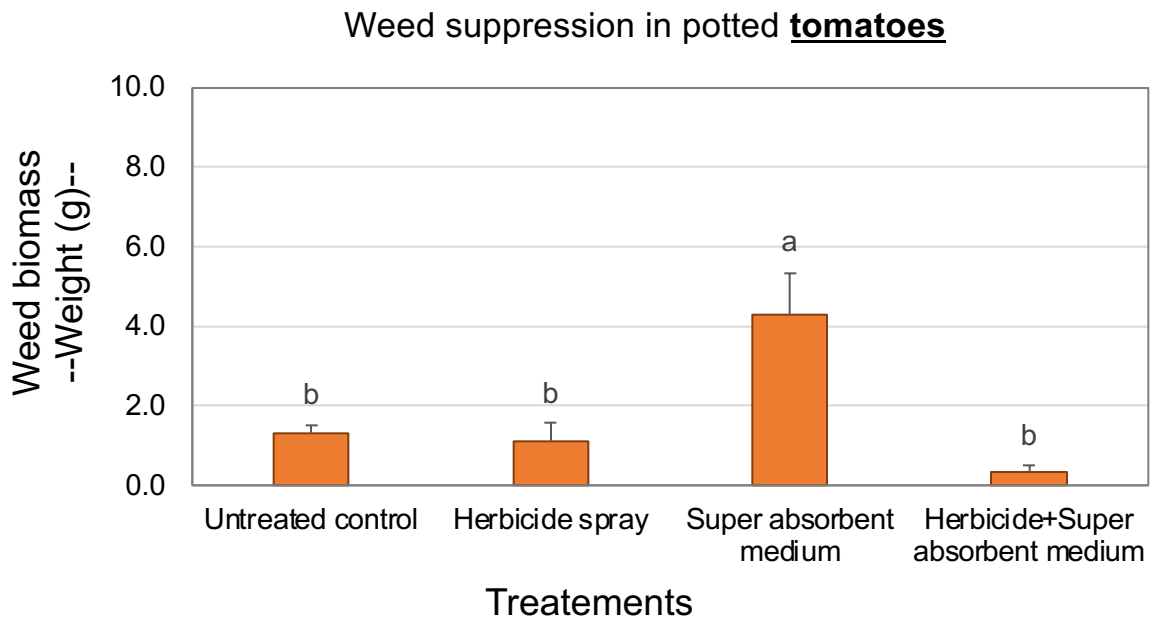


Figure 5. Effectiveness of treatments on suppressing weeds in pots planted with tomatoes. Biomass of weeds measured from the treated pots 4 weeks after planting. Bars with the same letters do not significantly differ based on Tukey's Honest Significant Difference (HSD) (α=0.05). Error bars indicate the standard error of 5 observations.

Table 1. Effects of treatments on the growth of plants in the pots

Treatments	Above ground biomass of plants in the planter pots[xy]	
	Onion	Tomato
Untreated control (UTC)	11.0 ± 4.46 ab[z]	18.2 ± 2.76 ab
Herbicide spray	4.3 ± 1.78 b	30.2 ± 1.81 a
Super absorbent medium only	14.7 ± 3.49 ab	28.3 ± 4.05 ab
Herbicide + super absorbent mixture	18.1 ± 3.36 a	12.7 ± 7.65 b

[x]Mean values ± Standard error; Number of observations: 5

[y]Biomass collected from the treated pots 4 weeks after planting

[z]Letters represent similarity within columns based on Tukey's Honest Significant Difference (HSD) (α=0.05)

Finding, Evaluating, and Fine-tuning Herbicide Alternatives to Glyphosate for the Florida Landscape Industry: Part II

PI: **Chris Marble, Environmental Horticulture, Mid-Florida REC**

ABSTRACT

Many naturally derived or organic non-selective contact herbicide options are available for landscape maintenance but not much research is available on the efficacy of these products. The objective of this research was to evaluate the efficacy of 9 different non-selective contact herbicides in comparison with glyphosate on common landscape weeds including doveweed (*Murdannia nudiflora*), yellow nutsedge (*Cyperus esculentus*), and longstalk phyllanthus (*Phyllanthus tenellus*). Overall, results were similar to previous research where glufosinate was the most effective herbicide and provided results similar to glyphosate on the two annual species evaluated (longstalk phyllanthus and doveweed). A very high level of control ("burndown") was noted at early evaluation dates with products including acetic acid, capric caprylic acid, pelargonic acid, ammoninum nonanoate and others, but plants recovered by the conclusion of the trial at 6 weeks after treatment. This indicates that many of these options may be effective but that sequential applications would be needed, and should most likely be targeted towards weeds at a younger growth stage than the mature plants that were evaluated in this trial.

OBJECTIVES AND METHODS

Due to negative publicity, many homeowners no longer want glyphosate to be used on their property and many municipalities in Florida have placed bans on glyphosate application on public property. As many landscape contractors rely on glyphosate to control troublesome and invasive weed species in landscapes, alternatives are needed in cases where glyphosate can no longer be used. In addition, regardless of bans or consumer preferences, alternative herbicides are needed for sound integrated weed management programs, to prevent/delay resistance development, and in many cases there are weeds for which glyphosate is not an effective option. While many non-selective alternatives exist, information is lacking on the efficacy of these herbicides against common weed species in Florida. Therefore, the objective of this research was to continue upon previous years' research and determine the efficacy of 9 different non-selective herbicides compared with glyphosate. In 2022, the weed species evaluated included longstalk Phyllanthus (*Phyllanthus tenellus*), doveweed (*Murdannia nudiflora*) and yellow nutsedge (*Cyperus esculentus*) (**Figure 1**).

Experiments were conducted at the Mid-Florida Research and Education Center in Apopka, FL in 2022. Nursery pots (1.9 liter) were filled with a pinebark:sand:peat potting soil that had been amended with a controlled release fertilizer [Osmocote Blend (8 to 9 month) 17-5-11] at a rate of 16 lbs. per cubic yard based on the manufacturer rate for incorporation. After filling, pots were moved to a full sun container pad and received 0.5 inches of overhead irrigation daily. On May 3, pots were seeded with approximately 30 seeds of either longstalk phyllanthus or doveweed while yellow nutsedge tubers were planted 1 inch deep at 5 tubers per pot. Weeds were allowed to grow until June 22 (~7 weeks after sowing, (**Figure 2**) when herbicide treatments (**Table 1**) were applied. Herbicides were applied using a CO_2 backpack sprayer calibrated to deliver 40 to 100 gallon per acre application volume (0.9 to 2.3 gal./1000 ft^2) based on herbicide label instructions (**Table 1**). Non-ionic

surfactants were added to each product if recommended on the manufacturer's label. Following treatment, pots were grouped by weed species in a completely randomized design with 8 single pot replications per treatment in each species. Data collected including visual control ratings at 1, 2, 4, and 6 weeks after treatment (WAT) on a 0 to 100 basis where 0 = no control or no difference with the non-treated pots (used for comparison) and 100 = 100% or no visible living plant tissues. At 6 weeks after treatment, shoot weight was assessed by clipping plants at the soil line and weighing on a portable field balance.

All data were subjected to analysis of variance using JMP statistical software (SAS) and arcsine transformed as needed to meet model assumptions. Each weed species was analyzed separately. For comparison purposes, all species and weed stages were also combined in order to compare the efficacy of each herbicide treatment over a range of species and growth stages. In all cases, treatment means were compared using Tukey's Honest Significant Difference Test and differences were considered significant at $P = 0.05$.

RESULTS

Yellow Nutsedge. At 1 WAT, the highest control was achieved with diquat followed by ammonium nonanoate with many other actives providing similar control such as acetic acid, caprylic + capric acid, and d-limonene (**Table 1**). Recovery was noted in almost all treatments by 2 WAT as expressed by decreasing control ratings. Exceptions included diquat, glyphosate, and glufosinate which either had similar or increased control ratings. This trend continued through 6 WAT with recovery noted in all treatments with the exception of glyphosate and diquat (**Figure 3**). Biomass data showed that while all treatments caused a reduction in yellow nutsedge growth, the best control was achieved with glyphosate and diquat which both provided a 100% reduction (or 100% control) in yellow nutsedge shoot biomass. It is important to note that this high level of control would not typically be expected with diquat. Diquat is a well-studied herbicide and has been researched much more thoroughly than other herbicides evaluated in these experiments. Diquat is a strictly contact herbicide and multiple applications are typically needed to control perennial weeds such as yellow nutsedge (see https://doi:10.1017/wet.2019.6 as an example). In this experiment however, 100% control was observed on all evaluation dates. It is unclear why such a high level of control was achieved which is in contrast to previous research. Additionally, when considering that all three weed species were grouped and treated together, it is unclear why diquat would provide complete control of nutsedge while providing only marginal control of phyllanthus and doveweed, both annual species. More research is needed but based on these results, most of the herbicides evaluated provided minimal to marginal suppression of nutsedge with one application.

Doveweed. At 1 WAT, the highest level of control was observed in plants treated with diquat or ammonium nonanoate, with both treatments providing approximately 80 to 90% control (**Table 2**). By 2 WAT, the highest control was achieved with glufosinate and diquat. Similar to results with yellow nutsedge, recovery was noted in most treatments throughout the remainder of the trial with the exception of plants treated with glyphosate or glufosinate which provided 66% control based on a visual control rating and a 72 to 73% reduction in doveweed shoot weight (**Figure 4**). Few other differences were observed among herbicides.

Longstalk Phyllanthus. Most herbicides evaluated provided a very high level of Phyllanthus control initially, with all herbicides with the exception of glyphosate and eugenol providing 60% control or higher (**Table 3**). This would be expected as Phyllanthus is typically an easier weed to control compared with doveweed and yellow nutsedge as it is an annual broadleaf weed which are typically more susceptible to non-selective contact herbicides. By 2 WAT recovery was noted in all treatments with the exception of plants treated with glyphosate (91% control). Ratings continued to decrease in all treatments throughout the remainder of the experiment (**Figure 5**). Based on biomass reduction, the highest level of control was achieved with glyphosate (87% reduction) and glufosinate (62% reduction) but herbicides including ammonium nonanoate, and pelargonic acid provided results similar to those observed with glufosinate.

CONCLUSIONS

When averaged across all three species, similar to 2020-2021 experiments, the best overall control was achieved with glyphosate (86%) followed by glufosinate (53%) and diquat which was similar to glufosinate (44%) (**Figure 6**). In almost all cases, significant recovery was noted at later weeks during the evaluation process, and was most notable from 2 WAT to 4 WAT. This could indicate that this 2 to 4 period would be the best ideal to make a sequential application to achieve satisfactory control. Determining the ideal re-application time with these treatments is currently our next and ongoing objective with this research. While formal control ratings were first conducted at 1 WAT, a very high level of visual control was noted with many of the contact products such as acetic acid, diquat, pelargonic acid, ammonium nonanoaate, and d-limonene, especially from 1 through 3 days after treatment. Further, many of these products provided very high levels of control at early evaluation dates (**Figure 7**). This indicates that these herbicides had significant but transient activity on these weeds as recovery was observed, at least on large weeds as were evaluated here. With the exception of glufosinate and in some cases diquat, few differences were noted among the other non-selective contact herbicides evaluated. For users wishing to utilize these herbicides, decisions could be based for the most part on worker safety (i.e. comparing PPE requirements, Danger vs. Caution labels, etc.), availability, price, and if other characteristics such as organic or OMRI (Organic Materials Review Institute) certification was important for them or their clients.

It should be noted that all three weed species were larger at the time of treatment than is typically recommended according to many of the product labels which recommend treating at a very young growth stage (e.g. 6 inches in height, 3 to 5 leaf, etc. or less). It would be expected that control ratings would be much higher if applications were made to weeds at an earlier timeframe relative to emergence than were evaluated here. Additionally, many of the products evaluated can be applied with a wide range of both concentrations and application volumes. Certain labels can be ambiguous with spot-spray rates and use verbiage such as "spray to wet" which can interpreted in different ways and lead to widely different application volumes (and consequently rates) being used in real-world situations. Therefore, we opted to choose broadcast rates (if provided), did not use the highest available application volume but chose 100 gallons/acre as the highest application volume (2.3 gal./1000 ft^2) evaluated, and we did not always choose the highest dose in all cases . Different results will likely be achieved with different rates, application volumes, and when applied to different weed species at different growth stages. This work is currently in the process of being replicated to validate results and further research is needed. Currently we are focusing our efforts on determining ideal reapplication timings and determining effective and realistic application volumes and rate concentrations to give Florida growers and landscape contractors the information they need to manage weeds in the most effective, economical, and environmentally friendly manner possible.

Table 1. Efficacy of glyphosate and alternative herbicides for control of yellow nutsedge (*Cyperus esculentus*) in Florida.

				7 Week Growth Stage[z]					
Treatment[y]				Visual Control Ratings (0 to 100)[x]				Biomass[w]	
Trade name	Herbicide (active)	Rate/acre	App. Vol.	1WAT	2WAT	4WAT	6WAT	F.W. (g)	% Control
Avenger Ag	d-limonene	20%	100	54 bcd	16 ef	6 de	11 b	40.0 ab	13 b
Axxe	Ammonium nonanoate	13%	100	70 b	29 cde	16 cde	11 b	39.7 ab	14 b
Terminator + Boost	Acetic acid	5%	100	61 bc	18 def	8 de	9 b	35.4 ab	23 b
Finale	Glufosinate	4 qt.	40	40 de	40 cd	25 cd	26 b	34.1 ab	26 b
FireWorxx	Caprylic + Capric acids	6% (v:v)	100	66 b	44 c	34 c	38 b	24.1 b	48 b
Ranger Pro	Glyphosate	2.66 qt.	40	30 e	68 b	74 b	98 a	0.2 c	100 a
Reward	Diquat	2 pts.	40	100 a	100 a	100 a	100 a	0.0 c	100 a
Scythe	Pelargonic acid	7% (v:v)	100	48 cde	23 cdef	10 de	33 b	29.7 ab	36 b
Green Gobbler	Acetic Acid	RTU	40	39 de	16 ef	6 de	9 b	37.4 ab	19 b
Weed Slayer	Eugenol	3 qt.	40	0 f	1 f	1 e	14 b	33.6 ab	28 b
Control	N.A.	N.A.	N.A.	---	---	---	---	46.2 a	---

[z]Growth stage shows the timing when herbicide applications were made relative to tuber planting.

[y]Rates are shown on a per acre basis. Application volume is shown in gallons per acre and was based on manufacturer label instructions.

[x]Visual control ratings were based on a 0 to 100 scale where 0 = 0% control or no difference from the non-treated control and 100 = 100% control or no visible green plant tissues present.

[w]Biomass shows shoot fresh weights collected at 6 weeks after treatment. % Control based on shoot weight reduction vs. control. Negative values indicate a percent increase relative to the non-treated control.

[v]Means followed by the same letter within each row and growth stage are not significantly different (Tukey's test, $P = 0.05$).

Table 2. Efficacy of glyphosate and alternative herbicides for control of doveweed (*Murdannia nudiflora*) in Florida.

				7 Week Growth Stage[z]					
Treatment[y]				Visual Control Ratings (0 to 100)[x]				Biomass[w]	
Trade name	Herbicide (active)	Rate/acre	App. Vol.	1WAT	2WAT	4WAT	6WAT	F.W. (g)	% Control
Avenger Ag	d-limonene	20%	100	68 bcd	21 c	9 bc	9 b	166.8 b	21 b
Axxe	Ammonium nonanoate	13%	100	79 ab	24 c	8 bc	8 b	169.0 ab	20 b
Terminator + Boost	Acetic acid	5%	100	54 e	23 c	3 c	4 b	221.0 a	-5 c
Finale	Glufosinate	4 qt.	40	60 cde	56 a	66 a	66 a	58.5 c	72 a
FireWorxx	Caprylic + Capric acids	6% (v:v)	100	58 de	30 c	0 c	1 b	197.0 ab	7 bc
Ranger Pro	Glyphosate	2.66 qt.	40	6 f	54 ab	55 a	66 a	56.2 c	73 a
Reward	Diquat	2 pts.	40	88 a	30 c	0 c	9 b	172.8 ab	18 bc
Scythe	Pelargonic acid	7% (v:v)	100	65 cde	34 bc	0 c	3 b	170.0 ab	19 bc
Green Gobbler	Acetic Acid	RTU	40	71 bc	14 c	0 c	0 b	220.9 a	-5 c
Weed Slayer	Eugenol	3 qt.	40	9 f	14 c	20 b	5 b	190.9 ab	9 bc
Control	N.A.	N.A.	N.A.	---	---	---	---	210.9 ab	---

[z]Growth stage shows the timing when herbicide applications were made relative to sowing seed.

[y]Rates are shown on a per acre basis. Application volume is shown in gallons per acre and was based on manufacturer label instructions.

[x]Visual control ratings were based on a 0 to 100 scale where 0 = 0% control or no difference from the non-treated control and 100 = 100% control or no visible green plant tissues present.

[w]Biomass shows shoot fresh weights collected at 6 weeks after treatment. % Control based on shoot weight reduction vs. control. Negative values indicate a percent increase relative to the non-treated control.

[v]Means followed by the same letter within each row and growth stage are not significantly different (Tukey's test, $P = 0.05$).

Table 3. Efficacy of glyphosate and alternative herbicides for control of longstalk phyllanthus (*Phyllanthus tenellus*) in Florida.

7 Week Growth Stage[z]									
Treatment[y]				Visual Control Ratings (0 to 100)[x]				Biomass[w]	
Trade name	Herbicide (active)	Rate/acre	App. Vol.	1WAT	2WAT	4WAT	6WAT	F.W. (g)	% Control
Avenger Ag	d-limonene	20%	100	93 a	45 cdef	11 cd	6 c	32.4 b	8 c
Axxe	Ammonium nonanoate	13%	100	99 a	55 bcde	25 bcd	25 c	25.8 bc	27 bc
Terminator + Boost	Acetic acid	5%	100	71 b	35 ef	0 d	5 c	35.1 ab	1 cd
Finale	Glufosinate	4 qt.	40	68 b	66 bc	61 ab	61 ab	13.5 cd	62 ab
FireWorxx	Caprylic + Capric acids	6% (v:v)	100	86 a	38 def	11 cd	14 c	31.1 b	12 c
Ranger Pro	Glyphosate	2.66 qt.	40	48 c	91 a	99 a	88 a	4.4 d	87 a
Reward	Diquat	2 pts.	40	89 a	60 bcd	20 cd	18 c	30.6 b	13 c
Scythe	Pelargonic acid	7% (v:v)	100	98 a	78 ab	40 bc	31 bc	25.3 bc	28 bc
Green Gobbler	Acetic Acid	RTU	40	60 bc	30 f	0 d	0 c	36.1 ab	-3 cd
Weed Slayer	Eugenol	3 qt.	40	4 d	0 g	0 d	0 c	46.2 a	-31 d
Control	N.A.	N.A.	N.A.	---	---	---	---	35.2 ab	---

[z]Growth stage shows the timing when herbicide applications were made relative to sowing seed.

[y]Rates are shown on a per acre basis. Application volume is shown in gallons per acre and was based on manufacturer label instructions.

[x]Visual control ratings were based on a 0 to 100 scale where 0 = 0% control or no difference from the non-treated control and 100 = 100% control or no visible green plant tissues present.

[w]Biomass shows shoot fresh weights collected at 6 weeks after treatment. % Control based on shoot weight reduction vs. control. Negative values indicate a percent increase relative to the non-treated control.

[v]Means followed by the same letter within each row and growth stage are not significantly different (Tukey's test, $P = 0.05$).

Figure 1. Species evaluated (L to R) included doveweed (Murdania nudiflora), longstalk Phyllanthus (Phyllanthus tenellus) and yellow nutsedge (Cyperus esculentus).

Figure 2. Yellow nutsedge, doveweed, and longstalk phyllanthus at the time of herbicide application.

Figure 3. Yellow nutsedge treatments (derandomized for photos) at 7 days (left) and 28 days (right) after herbicide treatment. Plants were treated (from L to R) with glyphosate, glufosinate, diquat, pelargonic acid, capric + caprylic acid, eugenol, acetic acid (Green Gobbler), acetic acid (Terminator), d-limonene, ammonium nonanoate, and nontreated.

Figure 4. Doveweed treatments (derandomized for photos) at 7 days (left) and 28 days (right) after herbicide treatment. Plants were treated (from L to R) with glyphosate, glufosinate, diquat, pelargonic acid, capric + caprylic acid, eugenol, acetic acid (Green Gobbler), acetic acid (Terminator + Boost), d-limonene, ammonium nonanoate, and nontreated.

Figure 5. Longstalk Phyllanthus treatments (derandomized for photos) at 7 days (left) and 28 days (right) after herbicide treatment. Plants were treated (from L to R) with glyphosate, glufosinate, diquat, pelargonic acid, capric + caprylic acid, eugenol, acetic acid (Green Gobbler), acetic acid (Terminator), d-limonene, ammonium nonanoate, and nontreated.

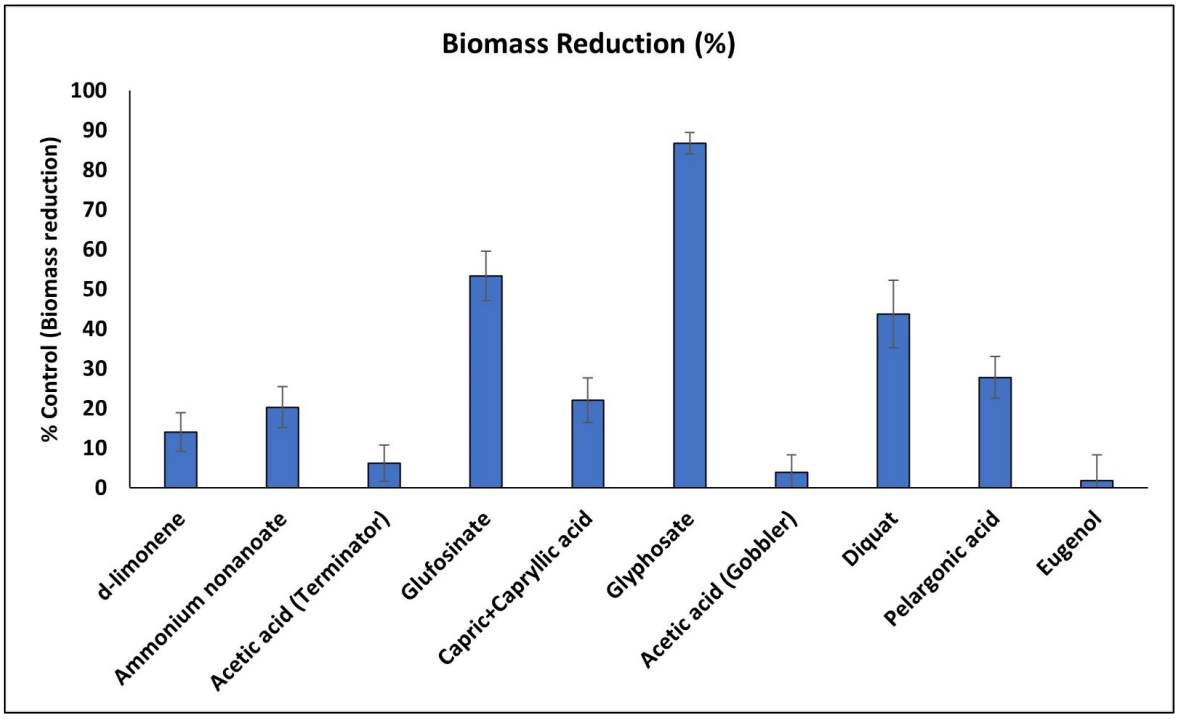


Figure 6. Mean % shoot weight (biomass) reduction (relative to non-treated) for 10 different herbicides averaged over results obtained from application to yellow nutsedge, longstalk Phyllanthus, and doveweed. Plants were harvested at 6 weeks after herbicides were applied.

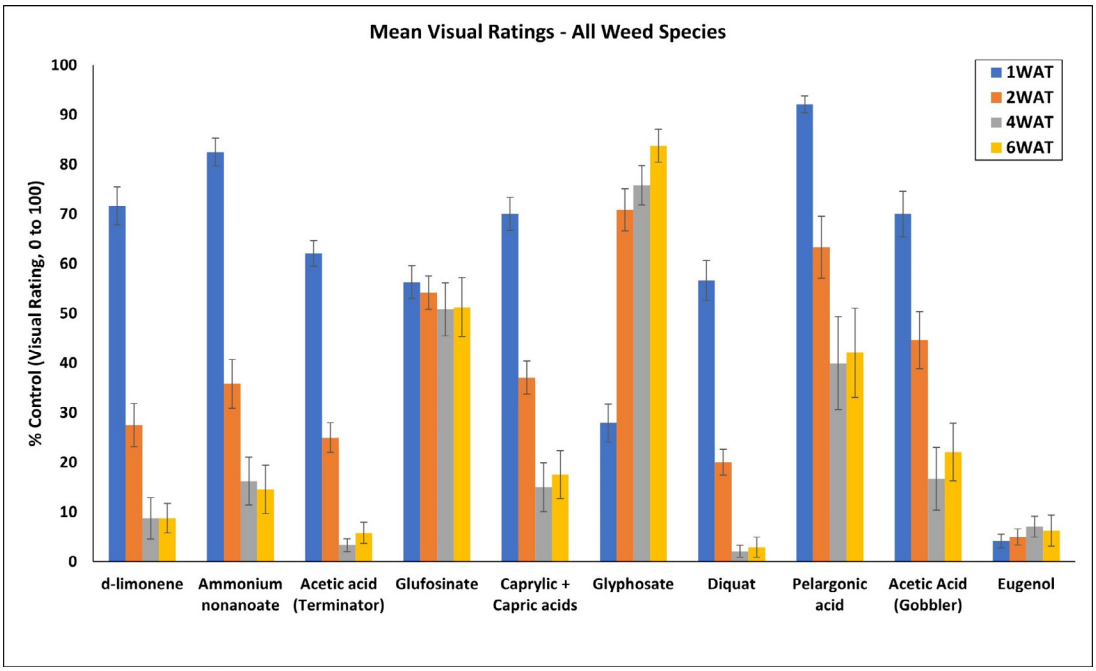


Figure 7. Visual control ratings taken on a 0 to 100% scale (0 = 0% control, 100 = 100% control/dead plant) at 1, 2, 4, and 6 weeks after herbicide treatment (WAT). Data shows means and standard errors for each herbicide over each rating period averaged over three different weed species including yellow nutsedge, longstalk phyllanthus, and doveweed.

Viburnum Foliar Disease Management: Disease mitigation during plant propagation

PI: **Gary Vallad, Plant Pathology, Gulf Coast REC**

Co-PIs: **Shawn Steed, Environmental Horticulture, UF/IFAS Extension &**
Wael Elwakil, Hillsborough County Extension

ABSTRACT

One of the top sellers of ornamental shrubs in Florida and placement in landscapes is the Viburnum species. This includes *Viburnum suspensum* and *V. odoratissimum*. Nurseries propagate Viburnum from using outdoor stock plants that may appear healthy, but often harbor several foliar diseases prevalent in nurseries and landscape environments. These diseases then compromise production during nursery production. This is especially problematic for ornamental shrub nursery production that relies on overhead irrigation, which is cheap to deploy but increases splash dispersal and leaf wetness that is optimal for plant disease spread and development, respectively. Most growers and landscapers struggle with disease control that reduces the salability of plants. While effective fungicide applications can help manage foliar diseases during container production, making applications during or immediately following propagation may improve liner production and limit disease losses following transplanting. We evaluated the benefit of applying effective fungicides during liner production as either a soil drench or as a dip-treatment for cuttings to minimize disease development early in the production cycle.

OBJECTIVES AND METHODS

Evaluate the benefit of applying fungicides during liner production to minimize disease dispersal and development early in the production cycle.

Methods: Cuttings of Sandankwa viburnum (*Viburnum suspensum*) were used to initiate two fungicide trials in the summer of 2022. Both trials were conducted in July thru August, using cuttings from naturally infected *Viburnum suspensum* plants that were grown in 3-gallon containers at a commercial production plant nursery in Hillsborough County. The trial was designed in completely randomized blocks with 5 replicates including a water control and five fungicide treatments. Fungicides were either applied as a soil drench (10 ml) to cuttings following the application of rooting hormone, or as dip treatment to prepared cuttings followed by the application of rooting hormone and planting. Per standard nursing practice, cuttings (12 per a replicate) were stuck in potting medium (50% peat: 50% perlite) on 6/21/22 in liner trays. Trays were immediately placed under an overhead mist system (4 seconds water cycle every 6 minutes) to keep soil moist. After an initial period of two weeks, cuttings were rated for disease incidence and severity on a weekly basis. The percentage of symptomatic foliage was rated weekly for six weeks to calculate the Area Under the Disease Progression Curve (AUDPC). Leaf tissues were periodically sampled for disease identification. After 6 weeks, plants were removed, given a final assessment on week 7 and uprooted to assess root length and root biomass (fresh and dry). Data analysis was conducted using a generalized mixed model analysis (PROC GLIMMIX) within SAS (version 9.4) with blocking as a random variable and fungicide treatment as a fixed effect, with the Student Panel command in SAS to check for normality. Disease severity over time was analyzed using the repeated measures function within GLIMMIX and a first-order autoregression to model covariance structure. Means separations were performed using Fisher's protected LSD at a 95% level of confidence and degree-of-freedom contrasts to further analyze differences between multiple treatment means.

RESULTS AND DISCUSSION

Identification of isolated fungi revealed the presence of multiple pathogens throughout both trials. Similar to previous nursery trials, *Colletotrichum* sp. was the most abundant, but others including *Cercospora* sp., *Corynespora* sp., and *Phyllosticta* sp. were observed causing symptoms of leaf spotting, blighting and defoliation on propagated cuttings. Both drench-applied and dip-applied fungicides had a statistically significant effect on disease based on final disease severity and AUDPC measurements (**Table 1** and **2**). For drench-applied fungicide treatments, both Postiva and Orkestra significantly reduced final disease severity by > 50% compared to the water control. In addition, both Postiva and Orkestra prevented disease severity from statistically increasing over time, relative to the control and other ineffective fungicide treatments (**Table 1**). For the drip-applied fungicide treatments, Postiva, Orkestra, Ryora, and Omega all significantly reduced final disease severity by 57% on average compared to the control. The same four treatments also prevented disease severity from statistically increasing over time, unlike the control. However, none of the treatments were statistically different from the control based on AUDPC.

Drench-applied fungicide treatments had a statistically significant effect on cutting vigor based on root length and root biomass (**Table 3**). Relative to the other fungicide treatments, Ryora resulted in cuttings with the shortest root length, and lowest fresh and dry root biomass compared to the control and the other fungicide treatments. Interestingly, dip-applied fungicide treatments did not have a statistically significant effect on cutting vigor (**Table 4**), and on average produced liner plants with roots that were on average 13% longer with 18% and 13% greater fresh and dry biomass, respectively, compared liner plants that received drench applied fungicide treatments.

CONCLUSIONS

Fungicides applied to propagative cuttings as either a dip treatment prior to planting or as a drench treatment following planting benefited disease control during liner production. Similar to nursery trials previously conducted, Orkestra and Postiva were the most effective at controlling the diverse diseases observed on the cuttings; whereas the other fungicide treatments were more variable depending on whether they were applied as a dip or drench. For example, Ryora and Omega, both appeared to perform better as a dip, rather than a drench. This would make sense for Omega (fluazinam), which is a contact fungicide, but Ryora (flutriafol) is a systemic fungicide. Topsin and Omega were included in these trials as broad-spectrum fungicides that have shown success against similar fungal pathogens on other crops. On average, drench-applied fungicide treatments appeared to have a larger impact on root development, including the controls that were treated with water. Fungicides can have profound impacts on plant growth. However, the observed differences in the water-treated controls suggests that the effect was likely due to the drench applications diluting the rooting hormone applied to the propagated cuttings prior to planting. Whereas in the other trial, the rooting hormone was applied after the cuttings were dipped in a fungicide solution.

Overall, while fungicide applications during propagation seems promising to control disease, it is not curative. We did not follow these studies through container production since disease severity at the end of both trials were similar to disease levels observed at the initiation of prior nursery trials. Hence, we don't believe the observed gains in disease control during these propagation trials are biologically significant given the favorable environmental conditions during subsequent container production due to the use of overhead irrigation. We propose developing alternative methods to propagate cuttings without the use of overhead irrigation, combined with use of drip-emitters and timely fungicide applications to minimize the impact on foliar diseases. In addition, the use of hot-water treatments, could be evaluated as a non-chemical means to cure cuttings of any latent infections.

Table 1. Effect of drench-applied fungicides to Viburnum cuttings for liner production on final disease severity, disease severity over time, and area under disease progression curve (AUDPC).

Product	Active Ingredient	FRAC	Rate/100 gal	P_{TIME}[Z]	DS_{final}[Y]	AUDPC[X]
Postiva	benzovindiflupyr + difenoconazole	7+3	28 fl oz	*0.0971*	5.8 c[W]	145 c
Orkestra	pyraclostrobin + fluxapyroxad	11+7	10 fl oz	*0.3392*	5.1 c	173 bc
Ryora	flutriafol	3	14 fl oz	*<.0001*	16.3 a	289 ab
Omega	fluazinam	29	16 fl oz	*<.0001*	16.8 a	317 a
Topsin 4.5L	Thiophanate methyl	1	20 fl oz	*0.0002*	9.5 bc	239 ab
Control (water)	-	-	- -	*<.0001*	12.7 ab	288 ab
			P-value =	*NA*	*0.0001*	*0.0618**

[Z] Statistical effect of fungicide treatments on disease severity over time, based on repeated measures.
[Y] Final disease rating, based on percent symptomatic foliage, collected on Aug. 19, 2022.
[X] Area Under the Disease Progression Curve (AUDPC), calculated using 7 disease severity ratings.
[W] Means followed by the same letter are not significantly different at the 95% level of confidence.
* *P-value* is of marginal significance, mean separation provided for guidance.

Table 2. Effect of dip-applied fungicides to Viburnum cuttings for liner production on final disease severity, disease severity over time, and area under disease progression curve (AUDPC).

Product	Active Ingredient	FRAC	Rate/100 gal	P_{TIME}[Z]	DS_{final}[Y]	AUDPC[X]
Postiva	benzovindiflupyr + difenoconazole	7+3	28 fl oz	*0.8919*	5.7 bc[W]	244 ab
Orkestra	pyraclostrobin + fluxapyroxad	11+7	10 fl oz	*0.9487*	4.1 c	131 b
Ryora	flutriafol	3	14 fl oz	*0.9585*	4.3 c	181 b
Omega	fluazinam	29	16 fl oz	*0.3415*	7.0 bc	236 ab
Topsin 4.5L	Thiophanate methyl	1	20 fl oz	*0.0775*	9.7 ab	325 a
Control (water)	-	-	- -	*0.0002*	12.3 a	243 ab
			P-value =	*NA*	*0.0011*	*0.0314*

[Z] Statistical effect of fungicide treatments on disease severity over time, based on repeated measures.
[Y] Based on final disease rating, based on percent symptomatic foliage, collected on Aug. 19, 2022.
[X] Area Under the Disease Progression Curve (AUDPC), calculated using 7 disease severity ratings.
[W] Means followed by the same letter are not significantly different at the 95% level of confidence.

Table 3. Effect of drench-applied fungicides to propagated Viburnum cuttings on final root length and fresh root and dry root biomass.

Product	Active Ingredient	FRAC	Rate/100 gal	*Root length (cm)*[Z]	Root biomass	
					Fresh (g)[Y]	Dry (g)
Postiva	benzovindiflupyr + difenoconazole	7+3	28 fl oz	4.8 a[W]	24.8 ab	3.8 a
Orkestra	pyraclostrobin + fluxapyroxad	11+7	10 fl oz	5.2 a	27.1 a	3.9 a
Ryora	flutriafol	3	14 fl oz	3.9 b	14.1 c	2.3 b
Omega	fluazinam	29	16 fl oz	5.0 a	19.0 bc	3.2 ab
Topsin 4.5L	Thiophanate methyl	1	20 fl oz	4.8 a	20.2 ab	3.2 ab
Water control	-	-	- -	4.9 a	22.3 ab	3.6 a
			P-value =	*0.0595**	*0.0372*	*0.0872**

[W] Means followed by the same letter are not significantly different at the 95% level of confidence.

* *P-value* is of marginal significance, mean separation provided for guidance.

Table 4. Effect of dip-applied fungicides to propagated Viburnum cuttings on final root length and fresh root and dry root biomass.

Product	Active Ingredient	FRAC	Rate/100 gal	*Root length (cm)*[Z]	Root biomass	
					Fresh (g)[Y]	Dry (g)
Postiva	benzovindiflupyr + difenoconazole	7+3	28 fl oz	5.7	30.2	4.4
Orkestra	pyraclostrobin + fluxapyroxad	11+7	10 fl oz	5.4	21.4	3.3
Ryora	flutriafol	3	14 fl oz	5.6	26.1	4.2
Omega	fluazinam	29	16 fl oz	5.7	29.5	4.1
Topsin 4.5L	Thiophanate methyl	1	20 fl oz	5.4	29.3	4.2
Water control	-	-	- -	5.7	18.9	2.7
			P-value =	*0.9479*	*0.1662*	*0.2210*

IMPROVE PRODUCTION SYSTEMS PRACTICES AND STRATEGIES

This priority area is defined as:

FNGLA supports research to develop advanced systems of product handling and transportation that will improve safety and efficiency.

FNGLA supported one project under this priority area, and the summary is on pages 45-48.

Improved Foliage Production Using Micropropagation - The Monstera Model

PI: **Wagner Vendrame, Environmental Horticulture**

ABSTRACT

Among foliage plants, Monstera is a genus of tropical evergreens very popular in the nursery industry. Rare and variegated varieties have a high value in the market. Micropropagation allows rapid clonal propagation of such varieties at a larger scale with the production of high quality and uniform plant material. Temporary immersion bioreactors (TIB) represent an advanced micropropagation system allowing increased plant multiplication and reduced costs. In this study, we evaluated the use of TIBs for the micropropagation of variegated Monstera 'Thai Constellation'. Plants grown in the greenhouse were disinfested and offshoots were removed and established in vitro using agar-based culture medium for multiplication. After multiplication in agar-based medium, five in vitro shoots were selected and established per bioreactor using liquid MS culture medium supplemented with 0.5 mg/L NAA and 7.5 mg/L BAP. Four immersion parameters were evaluated: immersion frequency every 1, 1.5 or 2 hours, and immersion duration for 1 or 2 minutes. Contamination was observed causing a delay in the experiment. Preliminary results indicate that immersion every 1.5 hour with a duration of 2 minutes is the best combination of parameters, providing an average of 6-8 shoots per initial explant. Additional bioreactors have been established and the study is under continuous evaluations to improve multiplication rates.

OBJECTIVES AND METHODS

1. To establish clean in vitro cultures of variegated Monstera using agar-based culture medium
2. To evaluate in vitro multiplication of Monstera in liquid culture medium using temporary immersion bioreactors
3. To evaluate temporary immersion parameters in bioreactors, including frequency and duration of immersion to optimize in vitro multiplication of Monstera

Methods

Plants of variegated Monstera 'Thai Constellation' (**Figure 1A**) were obtained from an ornamental nursery in South Florida and transferred to a greenhouse in the Department of Environmental Horticulture, IFAS, University of Florida, located in Gainesville, FL. Plants were maintained in the greenhouse and after 3 weeks, offshoots (**Figure 1B**) were removed from plants and transferred to laboratory to be used as explants. Surface disinfestation was performed using 1% Alconox detergent, after which the explants were submitted to deep disinfestation under laminar flow hood, consisting of 70% ethanol for 1 minute, 2% sodium hypochlorite for 20 minutes performed twice and three rinses in distilled autoclaved water.

Figure 1. Monstera 'Thai Constellation' plant (A). Offshoots used for in vitro studies (B).

Once disinfested, offshoot explants were split horizontally into two halves and established in vitro, with one half per container (baby-food jar) containing Murashige and Skoog (MS) culture medium. Supplemented with 0.5 mg/L NAA (1-naphthaleneacetic acid) and 5.0 mg/L BAP (6-benzylaminopurine). In vitro cultures were monitored weekly for growth and development, and contamination. Subculture to fresh culture medium was performed every 2 to 3 months. After 6 months, explants were transferred to fresh MS medium supplemented with 0.5 mg/L NAA and 7.5 mg/L BAP.

After 3 months, multiple shoots produced from explants were separated into individual shoots and transferred to temporary immersion bioreactors (TIBs) with 5 shoots per bioreactor. Immersion parameters were evaluated, including frequency of immersion (every 1, 1.5, and 2 hours) and immersion duration (1 and 2 minutes). Cultures were monitored weekly for multiplication, growth, and development. After 3 months, in vitro shoots produced in TIBs were transferred to the greenhouse for acclimatization.

RESULTS

Initially, contamination was a major obstacle to the establishment of in vitro cultures, delaying the study for a few months. Once the disinfestation protocol was optimized, clean in vitro cultures were easily established and showed stable growth and development. However, multiplication was slow under the initial agar-based MS medium containing 0.5 mg/L NAA and 5.0 mg/L BAP.

After 6 months, when cultures were transferred to fresh agar-based MS medium with 0.5 mg/L NAA and 7.5 mg/L BAP, multiplication increased with an average of 3-4 in vitro shoots produced per initial explant (**Figure 2A**). This initial multiplication phase under agar-based medium allowed the increase in plant material numbers for subsequent establishment of TIB studies.

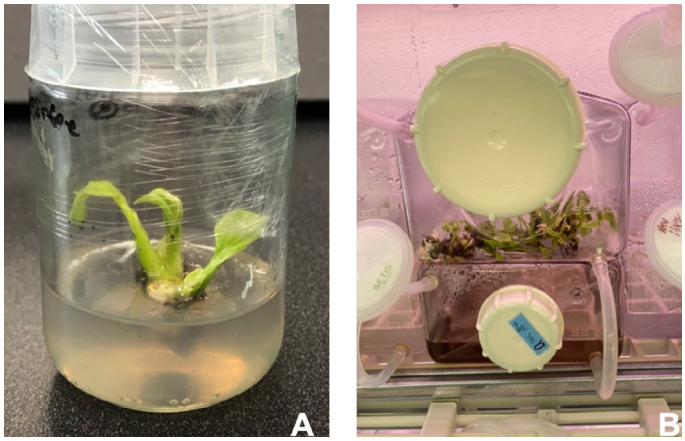


Figure 2. In vitro culture of Monstera in agar-based culture medium (A). Monstera shoots in temporary immersion bioreactors (B)

Subsequent transfers of in vitro shoots to bioreactors were successful and shoots showed stable growth and development (**Figure 2B**). Multiplication of in vitro shoots started at about 2 months after transfers to TIBs, faster as compared to agar-based medium. However, some contamination was observed, resulting from a leakage of air in the bioreactor system, thus causing a delay in the study. Once the problem was identified, it was promptly remediated and new cultures were established, with no signs of contamination.

Our preliminary results show that the best rate of multiplication was obtained under immersion frequency every 1.5 hour with duration of 2 minutes per immersion event in TIBs, with the production of 6-8 shoots per explant, thus doubling the rate of multiplication observed in agar-based systems. As the study is still under evaluations, we predict an increased multiplication rate in TIBs at a range of 3- to 4-fold the original multiplication rate in agar-based systems.

Some of the in vitro shoots produced in TIBs have since been rooted and transferred to greenhouse for acclimatization and further evaluation for survival, growth and development (**Figure 3**). To date, survival has been 100% with normal growth and development and no abnormalities have been observed.

Figure 3. In vitro-derived plantlets of Monstera in the greenhouse (A). Monstera transplanted to larger containers (B).

This study is still being conducted and will include experiments to improve the rates of multiplication by optimizing culture medium composition, such and combinations and concentrations of plant growth regulators, as well as the parameters for immersion frequency and duration.

CONCLUSIONS

Preliminary results from this study suggest that temporary immersion bioreactors can be a feasible system for the micropropagation of Monsteras and could serve as a model for other foliage ornamental crops. The rates of multiplication are higher compared to agar-based systems. In addition, the costs of production are significantly reduced when using bioreactors. The larger vessel size (4 L) compared to agar-based systems (100-200 ml) allows increased space and aeration as compared to agar-based systems, thus resulting in higher number of shoots produced per bioreactor unit. Furthermore, the use of liquid culture medium eliminates the need for agar, one of the most expensive elements of commercial micropropagation systems.

Future studies will incorporate other ornamental foliage plants and the evaluation of biostimulants to promote growth and development of in vitro-derived plants in the greenhouse during acclimatization.

GENETICS AND BREEDING TO ENHANCE QUANTITIES AND DIVERSITY OF PLANT MATERIAL

This priority area is defined as:

FNGLA supports research to improve the quality of plant material to improve ecological and social benefits.

FNGLA supported one project under this priority area, and the summary is on pages 50-54.

Developing a rapid molecular method to assess sugarcane mosaic virus (SCMV) load in turfgrass breeding materials

PI: **Jianping Wang, Agronomy**
Co-PIs: **Kevin Kenworthy, Agronomy & Philip Harmon, Plant Pathology**

ABSTRACT

Lethal Viral Necrosis (LVN) disease is caused by the sugarcane mosaic virus (SCMV). LVN is only known to occur in Floratam, the dominant St. Augustinegrass cultivar used in Florida landscapes. Floratam St. Augustinegrass planted in landscapes where the virus is present slowly progress from symptoms of mosaic disease to necrosis, specifically in winter. So far, there is no cure for the LVN disease. The goal of this project was to develop a rapid molecular method to assess SCMV load in turfgrass breeding materials, which thus will allow for determining the SCMV threshold in plants with LVN. Through the selection of specific primers to target SCMV sequence in turfgrass tissue and its application to qRT-PCR for viral detection this goal can be accomplished. In this project, the living green leaf tissue yielded the most concentrated and highest quality RNA extraction using the Direct-zol RNA Miniprep Kit (Zymo) and an additional Clean and Concentrator-5 Kit (Zymo). A specific primer targeting the SCMB coat protein and Nib gene with an amplificon size of about 2 kb was used to successfully amplify the viral genome from leave tissue of a St. Augustinegrass. The sequence was confirmed. The qRT-PCR was briefly conducted to determine the viral load and the experiment is still going on to be conclusive. The method will be further simplified for a rapid SCMV detection in the future.

OBJECTIVES

Sugarcane Mosaic Virus (SCMV) is a single-stranded, positive-sense RNA virus of the Potyvirus genus. The linear RNA genome is 9596 nucleotides surrounded by a non-enveloped capsid. In addition to its namesake Sugarcane plant, SCMV is also known to cause mosaic disease in turfgrasses throughout Florida starting in 2014. In the turfgrass cultivar Floratam St. Augustinegrass, Lethal Viral Necrosis (LVN) disease is caused by the sugarcane mosaic virus. LVN is only known to occur in Floratam, the dominant St. Augustinegrass cultivar used in Florida landscapes. Floratam St. Augustinegrass planted in landscapes where the virus is present slowly progress from symptoms of mosaic disease to necrosis, specifically in winter when temperatures drop below 65F. An entire lawn can become infected and die within a few years in addition to possibly infecting other lawns, thus the disease causes substantial economic losses. In contrast, when other St. Augustinegrass cultivars are infected by SCMV, only general mosaic chlorosis or yellow streaks develop on the leaves. So far, there is no cure for the LVN disease. Avoidance through sanitation practices, using resistant St. Augustinegrass cultivars or other species are the only options as it is a viral infection and cannot be controlled by fungicides or other pesticides.

The basis for Floratam's susceptibility is unknown. To breed alternative turfgrass with resistance to LVN, it is important to understand the mechanism of LVN development in Floratam St. Augustinegrass. Tissue sap of infected plants contains significant numbers of SCMV viral particles, which can inoculate uninfected plants, replicate, and travel through vascular into other plant tissues to cause different levels of symptoms depending on the host plant developmental stage and conditions.

Our hypothesis is that the SCMV replicates fast in Floratam St. Augustinegrass under low temperatures (<65F) and reaches a threshold level, which stimulates the necrosis symptom. However, in other cultivars or turf species, the SCMV replicates, but not at a rate to reach the threshold to cause plant necrosis. To test this hypothesis, we proposed to develop a quick real-time or quantitative reverse transcription PCR (qRT-PCR) method to specifically measure the SCMV load in Floratam St. Augustinegrass before and after cold treatment in comparison with other St. Augustinegrass cultivars, CitraBlue and Palmetto. qRT-PCR is a powerful tool for highly sensitive detection of plant viruses, which will provide a significant improvement over current immuno-assay technology. The SCMV genome sequences are readily available for PCR primer design, thus will facilitate the qRT-PCR method development.

The objective of this project is to develop a rapid molecular method to assess Sugarcane Mosaic Virus (SCMV) load in turfgrass breeding materials. Specifically, in Floratam St. Augustinegrass, the established method can then be utilized for determining the SCMV threshold in plants with Lethal Viral Necrosis (LVN). Through the obtention of plant viral RNA effectively, the selection of specific primers to target SCMV sequence in turfgrass tissue, and the application of said process to RT-qPCR for viral detection the development of a method to quickly and cost effectively assess SCMV load in infected turfgrass material is possible.

We will use a simplified method for obtaining plant viral RNA for RT-PCR analysis developed by Suehiro et al. 2005 from three leaf discs (a paper hole punch size), which takes about 15 min. Then we will combine the first-strand cDNA synthesis (RT step) and real time PCR as one step to increase the efficiency and reduce the error in the reaction (mimic the COVID 19 test). In developing this assay, we will design several degenerate primers according to the conserved regions of potyviruses and specific primers according to specific SCMV strain sequences. The efficiencies of these primers will be tested to choose the most efficient ones for the assay to assess either the total or specific strain of SCMV viral load of the infect plants.

Methods

This project utilized Sugarcane Mosaic Virus (SCMV) infected turfgrass material, the turfgrass cultivar presumed to be Floratam St. Augustinegrass. The turfgrass tissue material was symptom presenting including general mosaic chlorosis or yellow streaks on the leaves. The plant material was collected from the same grass stolon acquired from an infected lawn and provided by the turfgrass breeding program at the University of Florida and maintained in the UF greenhouse 676 off of 2350 Mowry rd.

Three tissue types, living root tissue, living green leaf tissue, and dead brown leaf tissue were collected and frozen in liquid nitrogen to preserve the integrity of the RNA as soon as the samples were cut from the plant. All the tissues were ground separately by mortar and pestle into fine powder. Each individual RNA extraction reaction included material from the same blade of grass.

Two extraction kits were compared during the RNA extraction process; RNeasy Plant Mini Kit (Qiagen) and Direct-zol RNA Miniprep Plus Kit (Zymo). Each kit's protocol was followed as directed. The Direct-zol kit included a DNase treatment, unlike the RNeasy kit. For this reason the samples were then processed further with RNA Clean and Concentrator-5 kit (Zymo) which has the option for a DNase treatment. This DNase treatment was applied to the RNeasy extraction in addition to the standard protocol.

To assess the quantity and quality of the RNA extracted, its concentration and purity was tested using Nanodrop. This provided a concentration (ng/uL) and a purity reading (260nm/280nm ratio; 260nm/230nm ratio) for comparing the two extraction methods. Ideal sample conditions include a high concentration and a purity ratio at or above 2.00 for each reading. The good quality and quantity RNA samples were reverse transcribed into cDNA using ProtoScript II First Strand cDNA Synthesis Kit (NEBiolabs).

A combination of designed primer pairs and existing primer pairs from literature were tested for best SCMV detection from the cDNA samples (**Table 1**). The primer distribution along the SCMV genome was show in **Figure 1**.

Primer criteria included specificity and exclusivity to the virus genome and the targeting of conserved SCMV genome regions which amplify a range of strains for broader detection.

Table 1. The sequences, amplicon size, location of the primers for amplifying the SCMV genome

Name	Sequence	Amplicon Size (bp)	Location (nt)
DP1 F	TTCGCCAAACACGACACCAC	1136	1835 – 2951
DP1 R	CAGTCCTCACTGCGCTAACT	1136	
DP2 F	AATGGAGGTGGCCAATCAGG	396	8553 – 8929
DP2 R	GTCTCCGTCCATCATCGTCC	396	
DP3 F	TGCCTCGTGTGTATGTGAGG	177	9405 – 9581
DP3 R	ACTCGCAGCACCAATGGTAA	177	
LP1 F	AGAGCACTTCAGCGTCATCAGA	1711	5829 – 7539
LP1 R	CAAACGGTTCCACCCACCATAA	1711	
LP2 F	GACTCGGACTTTTACAGCAG	1966	7392 – 9357
LP2 R	AACAGGGTTTCCAGGAGACT	1966	
LP3 F	CATGTTGCTGCGTTACAACGG	1509	1724 – 3232
LP3 R	TTGCTTCAATGAGGCGTGGGT	1509	

Note: Primers selected for further testing. Designed Primers (DP) were designed using NCBI Primer Blast Tool. Literture Primers (LP) were extracted from: Xie et. Al., 2016. *Molecular Variability and Distribution of Sugarcane Mosaic Virus in Shanxi, China*. PLoS One. 11(3): e0151549.

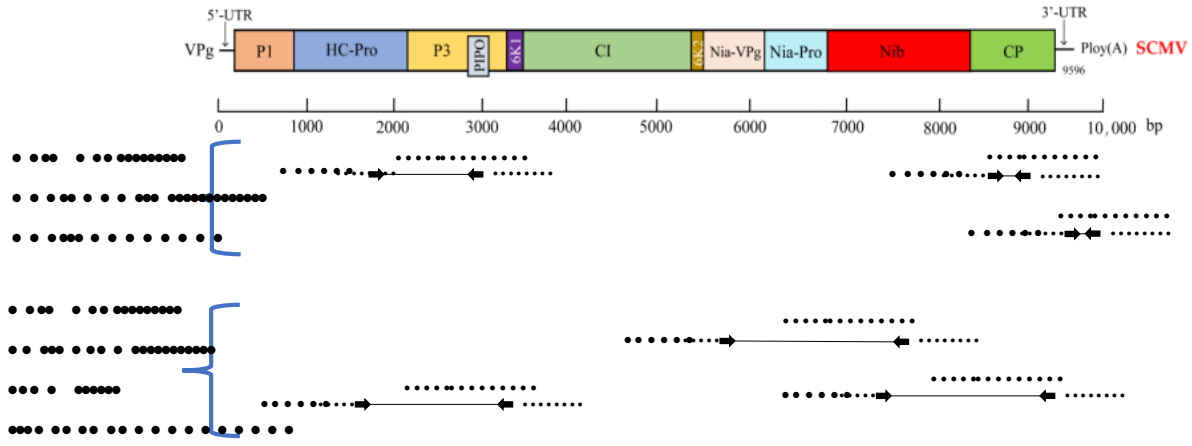


Figure 1. the primer distribution along the SCMV genome (top). DP1, DP2, DP3 are designed in our lab. LP1, LP2, and LP3 are according to Xie et. Al., 2016.

Initial testing conditions for each of the primers included running each primer on an annealing temperature gradient to find the optimal temperature. The gradients were based on the primer pair's melting temperature (TM-5*C) and provided a range surrounding that point. Further optimization was done through more stringent reaction conditions such as higher annealing temperatures and variations in concentrations of primer and template. Additionally, a range of DNA Polymerases were tested for most efficient PCR results. Primers chosen for the next step of the project were those which successfully amplified their designed target. This was confirmed through 1% agarose gel electrophoresis at 90 V for 40 minutes and subsequent Sanger Sequencing of the specific amplicons.

qRT-PCR was conducted by using the KAPA Sybr Fast Universal kit following the manufacturer's instruction with ubiquitin as the internal control.

RESULTS

Of the three tissue types, living green leaf tissue, living root tissue, dead brown leaf tissue, living green leaf tissue provided the highest concentration and quality of RNA. Living green leaf tissue rendered the highest concentration in both RNA extraction kits (RNeasy Plant Mini Kit (Qiagen) and Direct-zol RNA Miniprep Plus Kit (Zymo)) when measured using Nanodrop, meanwhile the living root tissue rendered the least RNA with the lowest quality extraction. Dead brown leaf tissue provided high concentrations of RNA but lower purity extractions.

Table 2. The RNA sample concentration and quality measurements isolated by using different kits from different tissue types.

Sample	ng/uL	260nm/ 280nm	260nm/ 230nm
Direct-zol Extraction Green Leaf + Clean and Concentrator-5 Kit	466.75	2.10	2.23
Direct-zol Extraction Brown Leaf + Clean and Concentrator-5 Kit	97.21	2.18	1.99
Direct-zol Extraction Root + Clean and Concentrator-5 Kit	1.44	1.85	0.55
RNeasy Extraction Green Leaf + Clean and Concentrator-5 Kit	116.94	2.15	1.12
RNeasy Extraction Brown Leaf + Clean and Concentrator-5 Kit	121.47	2.17	0.81
RNeasy Extraction Root + Clean and Concentrator-5 Kit	11.44	2.17	1.20
RNeasy Extraction Green Leaf + Clean and Concentrator-5 Kit + DNase treatment	31.01	2.06	0.99
RNeasy Extraction Brown Leaf + Clean and Concentrator-5 Kit + DNase treatment	28.26	2.10	0.65
RNeasy Extraction Root + Clean and Concentrator-5 Kit + DNase treatment	3.33	1.32	0.83

Note: Nanodrop results for the three tissue types tested with two RNA extraction kits and further processed with a Clean and Concentrator-5 kit. The RNeasy extractions received additional DNase treatment as it was not included in the RNeasy kit.

When two RNA extraction kits utilized were compared, the Direct-zol RNA Miniprep Kit rendered the highest quality extractions and already included the DNase treatment unlike the RNeasy Plant Mini Kit. For this reason, the RNA utilized for the remainder of the project was extracted using Direct-zol RNA Miniprep Kit with the Clean and Concentrator-5 Kit.

Following primer testing, only 3 of the 6 primer pairs (**Table 1**) selected showed promising results (**Figure 2**). Primer LP1 showed little to no amplification during the gradient annealing temperature tests and was dropped from further testing. Primers DP1 and LP3 did not generate the desired amplicon size and appeared to have off target matches, these two were also dropped. Primers DP2 and DP3 amplified at their respective amplicon size and were being optimized to amplify a singular band. Primer LP2 was successfully optimized, and the PCR product was confirmed by Sanger Sequence (**Figure 3**). Currently the qRT-PCR experiment is going on for viral load detection.

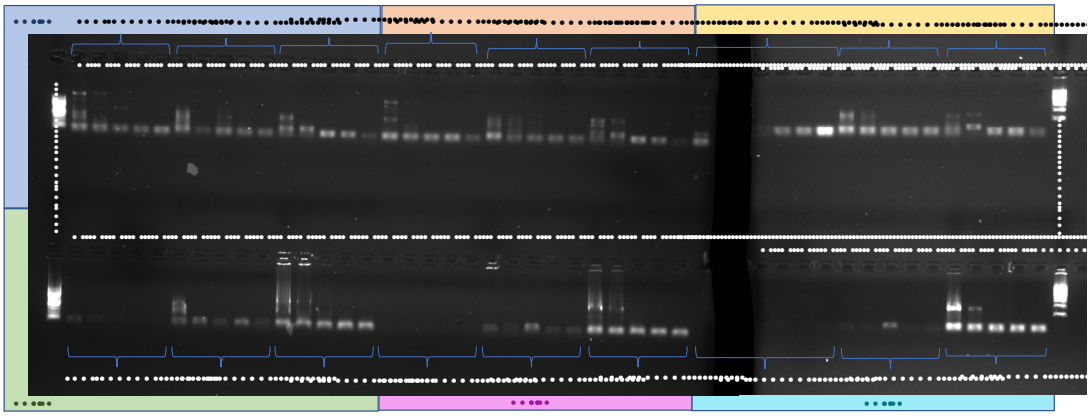


Figure 2. The Example gel of primer optimization at different annealing temperatures.

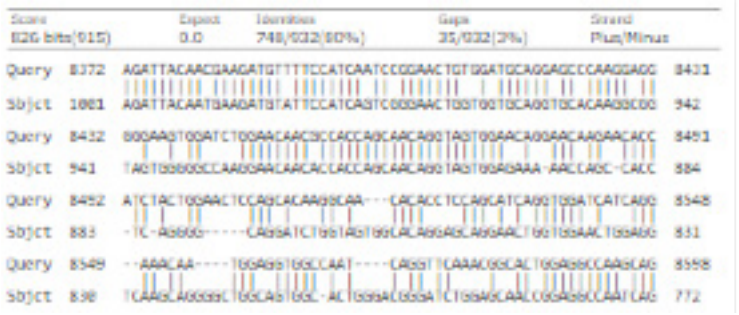


Figure 3. Sanger sequencing confirmation of the amplicon (example of the sequence alignment between the SCMV genome and the amplicon sequence).

CONCLUSION

The establishment of a method to quickly and cost effectively assess SCMV load in infected turfgrass material using plant viral RNA would assist in the selection of breeding materials in the effort to develop St. Augustinegrass cultivars free or having low viral infection. Through the selection of specific primers to target SCMV sequence in turfgrass tissue and its application to qRT-PCR for viral detection this goal can be accomplished. RNA samples were successfully extracted from living green and brown leaf tissue of infected St. Augustinegrass and the SCMV genome was specifically detected through RT-PCR by using two pairs of primers targeting the SCMV genome. The qRT-PCR method is currently used to detect the viral load, which will be further simplified for a rapid SCMV load detection in turfgrass breeding materials.

List of FNGLA Funded Projects Since 2005-06

2005-2006

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Thomas Yeager	Environmental Horticulture	Gainesville Campus	Statewide Expansion of South Florida BMP Effort
William Crow	Entomology & Nematology	Gainesville Campus	Biological Control of Root-Knot Nematodes on Woody Ornamentals
Forrest Howard	Environmental Horticulture	Ft. Lauderdale REC	Biology and Management of West Indies Mahogany Scale, Conchaspis cordiae (Hemiptera: Conchaspididae)
Zhanao Deng	Environmental Horticulture	Gulf Coast REC	Genetic Sterilization of Lantana
David Clark	Environmental Horticulture	Gainesville Campus	Development of New Coleus Cultivars for Better Foliage Color Stability and Use as Groundcovers
James Gibson	Environmental Horticulture	West Florida REC	Consumer Purchase Patterns in Florida (3-year study) Study 1 (completed): The Impact of In-House Displays on Impulse Buying Behavior; Study 2 (ongoing project): The Impact of Display Gardens on Identifying Consumer Needs, Trends, and Preferences; Study 3: (Proposed): Developing Employee Plant Knowledge to Effectively Educate Consumers and Increase Sales

2006-2007

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
James Barrett	Environmental Horticulture	Gainesville Campus	Evaluating Flowering Annuals and Herbaceous Perennials for the Florida Climate
Monica Elliott	Plant Pathology	Ft. Lauderdale REC	Determine the etiological agent for a new disease affecting Syagrus romanzoffiana (queen palm) in landscapes and nurseries
Kati Migliaccio	Agricultural & Biological Engineering	Tropical REC	Designing Irrigation BMPs Considering Capillary Rise for Production Cost Savings
Kimberly Moore	Environmental Horticulture	Ft. Lauderdale REC	Fertilization Effects on Water Requirements of Container Grown Ornamentals during Establishment in the Landscape
Wagner Vendrame	Environmental Horticulture	Tropical REC	Potential Horticultural and Disease Management Benefits of Silicon Fertilization of Potted Orchids
Tom Yeager	Environmental Horticulture	Gainesville Campus	Expanded BMP Education

2007-2008

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Kimberly Moore	Environmental Horticulture	Ft. Lauderdale REC	Organic Matter and Irrigation Frequency Effects During Shrub Establishment
Tom Yeager	Environmental Horticulture	Gainesville Campus	BMP Workshops for Field-Grown Plant Producers
Michael Dukes	Agricultural & Biological Engineering	Gainesville Campus	Development of Programming Recommendations for Smart Irrigation Controllers
Gurpal Toor	Soil & Water Sciences	Gulf Coast REC	Characterization of Organic Compounds in Nursery Reclaimed Water
Monica Elliot	Plant Pathology	Ft. Lauderdale REC	Fusarium Decline of Palms: Pathogen, Hosts, Diagnosis and Control
Zhanao Deng	Environmental Horticulture	Gulf Coast REC	Toward Sterilizing Nandina: Inducing Tetraploids for Development of Sterile, Non-Invasive Triploid Nandina
Francisco Escobedo	School of Forest Resources & Conservation	Gainesville Campus	The Benefits of Florida's Urban Forests on Environmental Quality

2008-2009

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Richard Beeson	Environmental Horticulture	Mid-Florida REC	Commercial Evaluation of Automated Irrigation Control for Overhead Irrigation Based on Daily Weather
Geoffrey Denny	Environmental Horticulture	Gulf Coast REC	Validation of Nitrogen Fertilizer Recommendations for Florida Landscape Plants
Michael Dukes	Agricultural & Biological Engineering	Gainesville Campus	Irrigation Controller Programming Guidelines by Multimedia Methods
Paul Fisher	Environmental Horticulture	Gainesville Campus	Onsite Monitoring of Water Treatment Technologies in Recycled Irrigation Water for Florida Nurseries
Paul Monaghan	Agricultural & Biological Engineering	Gainesville Campus	Using Community Based Social Marketing to Evaluate Homeowner Attitudes Towards Florida Friendly Waterfront Landscapes
Brian Pearson	Environmental Horticulture	Mid-Florida REC	Quantification of Stormwater Nutrient Runoff in the Environment
Amy Shober	Soil & Water Sciences	Gulf Coast REC	Effects of Organic Matter and Tillage on Plant Establishment and Nutrient Losses in an Residential Landscape
Thomas Yeager	Environmental Horticulture	Gainesville Campus	Production Strategies for Water Savings in the Landscape

2009-2010

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Jianjun Chen	Environmental Horticulture	Mid-Florida REC	Improving the Quality of Recycled-Irrigation Water by Minimizing Algal Density Using Plant-Friendly Chemicals
Geoffrey Denny	Environmental Horticulture	Gulf Coast REC	Validation of Nitrogen Fertilizer Recommendations for Florida Landscape Plants
Rosanna Freyre	Environmental Horticulture	Gainesville Campus	Breeding of Sterile and Non-Invasive Ruellia Cultivars
Jason Keith Kruse	Environmental Horticulture	Gainesville Campus	Determining Required Width of Unfertilized Buffer Strips to Limit Fertilizer Movement Into SurfaceWater Bodies
Amy Shober	Soil & Water Sciences	Gulf Coast REC	Evaluation of Soil Physical and Chemical Properties at Newly Constructed Residential Home Sites to Improve Plant Growth and Environmental Quality
Tom Yeager	Environmental Horticulture	Gainesville Campus	Developing a BMP Manual for Field-Grown Plant Producers
Tom Yeager	Environmental Horticulture	Gainesville Campus	Automatic Irrigation Control Based Upon Plant Need

2010-2011

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
David Clark	Environmental Horticulture	Gainesville Campus	The University of Florida Sensory Gardens
Catharine Mannion	Entomology & Nematology	Tropical REC	Impact of Insecticides and Method of Application on Natural Enemies in the Landscape
Kimberly Moore	Environmental Horticulture	Ft. Lauderdale REC	Use of Reclaimed Waste Water to Grow Greenhouse Ornamental Plants
Kati Migliaccio	Agricultural & Biological Engineering	Tropical REC	Interactive Tool for Improving Water Management in Landscapes
Robert Stamps	Environmental Horticulture	Mid-Florida REC	Evaluation and Identification of Effective and Safe Herbicides, Herbicide Formulations and Application Rates for Landscape and Nursery Use
Tom Yeager	Environmental Horticulture	Gainesville Campus	Development of an Economic Decision Support Tool for Container Nursery Management
Tom Yeager	Environmental Horticulture	Gainesville Campus	Enhanced Decision Capabilities for Irrigation of Container Plants

2011-2012

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Gul Shad Ali	Plant Pathology	Mid-Florida REC	Development of a Rapid and Sensitive Diagnostic Kit for Ornamental Plant Pathogens Using Loop-Mediated Isothermal Amplification and Recombinase Polymerase Amplification
Erin Alvarez	Environmental Horticulture	Gainesville Campus	The University of Florida Sensory Gardens
Eileen Buss	Entomology & Nematology	Gainesville Campus	Gall-Maker Management in Live Oak Nurseries
Aaron Palmateer	Plant Pathology	Tropical REC	Management of High Consequence Bacterial
Amy Shober	Soil & Water Sciences	Gulf Coast REC	Evaluation of Nutrient Leaching From Mixed Landscapes
Tom Yeager	Environmental Horticulture	Gainesville Campus	Continued Development of an Economic Decision Support Tool for Container Nursery Management

2012-2013

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Tom Yeager	Environmental Horticulture	Gainesville Campus	Evaluating the Effect of Plant Species on Water Usage to Improve Container Nursery Irrigation BMPs
James P. Cuda	Entomology & Nematology	Gainesville Campus	Mass Rearing of the South American Psyllid Calophya terebinthifolii (Hemiptera: Calophyidae), a Candidate Biological Control Agent for Brazilian Peppertree
Gary Knox	Environmental Horticulture	North Florida REC	New Crapemrytle Cultivars for the Southeastern U.S. An Extensive Evaluation of Field Resistances to Fungal, Bacterial and Abiotic Disorders and Plant and Flower Characteristics
Tesfamariam Mengistu	Entomology & Nematology	Gainesville Campus	Development of a New Molecular Method to Detect Major Root-Knot Nematodes (Meloidogyne spp.) Occurring in Florida Nurseries
Gul Shad Ali	Plant Pathology	Mid-Florida REC	Implementation and Field Testing of a Rapid and Sensitive Diaignostic Kit for Ornamental Plant Pathogens Using Loop-Mediated Isothermal Amplification Integrated with Lateral Flow Devices
Monica Elliott	Plant Pathology	Ft. Lauderdale REC	Fungicide Movement, Distribution and Persistence in Palms
Robert Stamps	Environmental Horticulture	Mid-Florida REC	Development of Control and Eradication Methods for a Weed Posing a Nursery Quarantine Risk and a Weed Posing Human Health and Environmental Risks
Zhanao Deng	Environmental Horticulture	Gulf Coast REC	Developing Superior Native Plant Varieties for the Florida Nursery and Landscape Industry

2013-2014

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Steven Arthurs	Entomology & Nematology	Mid-Florida REC	Processed Coffee Grounds to Manage Cycad Aulacaspis Scale in Landscapes
Jianjun Chen	Environmental Horticulture	Mid-Florida REC	Developing Color-Leaved Ficus Plants Through Biotechnology Approaches
Huangjun Lu	Horticultural Sciences	Everglades REC	Enhancing St. Augustinegrass for Drought Tolerance
Paul Monaghan	Agricultural & Biological Engineering	Gainesville Campus	Increasing Tree Sales and Survivability in Urban Areas Community Tree Stewardship Programs
Kimberly Moore	Environmental Horticulture	Ft. Lauderdale REC	Determination of Salt Tolerance of Container Grown Ornamental Shrubs
Quisto Settle	Agricultural & Biological Engineering	IFAS Center for Public Issues Education	Understanding Public Opinion of Issues Facing the Nursery and Landscape Industry in Florida
Thomas Yeager	Environmental Horticulture	Gainesville Campus	Enhancing Irrigation in Container Nurseries Using Mobile Device App
Thomas Yeager	Environmental Horticulture	Gainesville Campus	Develop Video to Promote BMPs

2014-2015

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Bala Rathinasabapathi	Horticultural Sciences	Gainesville Campus	Toward a Novel Biopesticide to Control Fall Armyworms: Beebalm Phytochemicals
Tom Yeager	Environmental Horticulture	Gainesville Campus	A Mobile Device App for Enhancing Irrigation in Container Nurseries
Aaron Palmateer	Plant Pathology	Tropical REC	Using Plant Diagnoistic Reports as a Tool for Preventative Disease Management in Florida Nurseries and Landscapes
Ronald Cave	Entomology & Nematology	Indian River REC	Biological Control of Green Croton Scale on Ornamental Plants
Stephen Marble	Environmental Horticulture	Mid-Florida REC	Increasing the Accuracy and Effectiveness of Herbicide Applications in Florida Nurseries
Mathews Paret	Plant Pathology	North Florida REC	Rose Mosaic: Management of Destructive Rose Virus Complex Using Early Detection and Novel IPM Strategies
Nathan Boyd	Horticultural Sciences	Gulf Coast REC	Weed Management Options for Tropical Ornamentals
Erica Goss	Plant Pathology	Gainesville Campus	New Method to Detect Hybrid Phytophthora in Nursery Production
Catharine Mannion	Entomology & Nematology	Tropical REC	Contributing Factors in Ficus benjamina Decline

2015-2016

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Mace Bauer	Horticultural Sciences	Gainesville Campus	Improve Environment and Resource Management
Nathan Boyd	Horticultural Sciences	Gulf Coast REC	Weed Management Options for Tropical Ornamentals
Paul Fisher	Environmental Horticulture	Gainesville Campus	Delivering Adequate Oxygen for Rooting of Plant Cuttings
Paul Fisher	Environmental Horticulture	Gainesville Campus	Lowcost and Automated Sensorbased Technology for Improving Irrigation Strategies
Stephen Marble	Environmental Horticulture	Mid-Florida REC	Determining the Impact of Metsulfuron a Turf Herbicide on Growth and Establishment of Ornamental Trees and Shrubs in Florida's Landscapes
Kimberly Moore	Environmental Horticulture	Ft. Lauderdale REC	Varying Leaching Fractions and Waste Water Blends to Grow Containerized Foliage Plants
Bart Schutzman	Environmental Horticulture	Gainesville Campus	Expansion and Enhancement of the Gardens at Fifield for Research, Teaching and Extension
Tripti Vashisth	Horticultural Sciences	Citrus REC	Evaluate the Use of Plant Growth Regulators and Different Growing Media to Accelerate the Rate of Germination and Growth in Citrus Rootstock Seedlings and Budded Trees
Tom Yeager	Environmental Horticulture	Gainesville Campus	Using Leaching Fraction to Achieve Appropriate Irrigation Application Amounts

2016-2017

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Brian Bahder	Entomology & Nematology	Ft. Lauderdale REC	Evaluation of Insects in Areas Impacted by Texas Phoenix Palm Decline for Their Potential as Vectors
Nathan Boyd	Horticultural Sciences	Gulf Coast REC	Preemergence Herbicides for Weed Control in Allamanda,Bird of Paradise, Firebush and Hibiscus
Adam Dale	Entomology and Nematology	Gainesville Campus	Novel Cultural Strategies for Managing Insect Pests of St. Agustinegrass
Paul Fisher	Environmental Horticulture	Gainesville Campus	Remediating Agrichemicals from Irrigation Water Using an Activated Carbon Filter
Rosanna Freyre	Environmental Horticulture	Gainesville Campus	Breeding Sterile Dwarf Mexican Petunia (Ruellia Simplex) at the University of Florida
Catharine Mannion	Entomology and Nematology	Tropical REC	Managing Ficus Whitefly Without Pesticides
S. Chris Marble	Environmental Horticulture	Gainesville Campus	Impact of Herbicide Application Carrier Volume on Weed Control in the Absence of Rainfall or Irrigation for Activiation
Xavier Martini	Entomology and Nematology	North Florida REC	Investigating Potential Alternative Vectors and Reservoirs of Rose Rosette Virus in the Florida Panhandle
Bryan Unruh	Environmental Horticulture	West Florida REC	A Mobile Web Application for Geolocating Fertilizer Ordinance Jurisdictions

2017-2018

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Charles Guy	Environmental Horticulture	Gainesville Campus	Assessing Human Health Benefits of Gardening
Raymond Odeh	Environmental Horticulture	Gainesville Campus	
Allan Bacon	Soil and Water Science	Gainesville Campus	Long-term Recovery of Compacted Residential Soils
Eben Broadbent	Forest Resources and Conservation	Gainesville Campus	
Adam Dale	Entomology and Nematology	Gainesville Campus	Investigating the Causal Agent of Bud Galls on Florida Ornamental Plants
Gul Shad Ali	Plant Pathology	Mid-Florida REC	
Erin Harlow	Duval County Extension	IFAS Extension	
Rhuanito Ferrarezi	Horticultural Sciences	Indian River REC	Accelerated Production of Citrus Nursery Trees Using Automated Ebbandflow Subirrigation
Basil Iannone	Forest Resources and Conservation	Gainesville Campus	Planting Stormwater Ponds: Determining the Benefits and Best Management Practices for Ornamental Plants in an Underutilized Portion of Residential Landscapes
Michelle Atkinson	Manatee County Extension	IFAS Extension	
Mary Lusk	Soil and Water Science	Gulf Coast REC	
Tom Yeager	Environmental Horticulture	Gainesville Campus	Redefining Irrigation Permit Allocations for Nurseries
Brian Bahder	Entomology and Nematology	Ft. Lauderdale REC	Developing dPCR for Detecting Phytoplasmas in Palms
Heqiang "Alfred" Huo	Environmental Horticulture	Mid-Florida REC	Development of Genetically Engineered Banker Plants for Biological Control of Whiteflies in Greenhouses
Lance Osborne	Entomology and Nematology	Mid-Florida REC	
H. Dail Laughinghouse	Agronomy	Ft. Lauderdale REC	Developing Effective Management Options for *Nostoc* spp. in Florida Nurseries
Chris Marble	Environmental Horticulture	Mid-Florida REC	
David Berthold	(No Unit Affiliation)	Ft. Lauderdale REC	
Mathews Paret	Plant Pathology	North Florida REC	Recent Widespread Damage of Commercial and Landscape Roses In Florida To Crown Gall Disease: Characterizing the Bacterial Strains and Establishing Management Strategies
Gary Knox	Environmental Horticulture	North Florida REC	

2018-2019

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Andrew Koeser	Environmental Horticulture	Gulf Coast REC	Determining Root Space Requirements for Florida Street Trees
Deb Hilbert	Environmental Horticulture	Gulf Coast REC	
Heidi Radunovich	Family, Youth and Community Sciences	Gainesville Campus	Identifying the Impacts of Opioids on Florida Nursery, Growers and Landscapers
Christa Court	Food and Resource Economics	Gainesville Campus	
Heqiang "Alfred" Huo	Environmental Horticulture	Mid-Florida REC	Development of Salinity Tolerant Petunia Through CRISPR/Cas9 GeneEditing
Linhchi Nguyen	Environmental Horticulture	Mid-Florida REC	
Tom Yeager	Environmental Horticulture	Gainesville Campus	Use of Reclaimed Water in Production Nurseries
Shawn Steed	Hillsborough County Extension	IFAS Extension	
Brian Bahder	Entomology and Nematology	Ft. Lauderdale REC	Evaluating vector potential of Haplaxius crudus and Idioderma virescens
Thomas Chouvenc	Entomology and Nematology	Ft. Lauderdale REC	Measuring the Impact of a New Invasive Ant Species (Plagiolepis alluaudi) on Plant Feeding Insects in South Florida Nurseries
Brian Bahder	Entomology and Nematology	Ft. Lauderdale REC	
Andrea Lucky	Entomology and Nematology	Gainesville Campus	
Chris Marble	Environmental Horticulture	Mid-Florida REC	Improving Nursery Weed Control by Choosing Herbicides Based on Application Timing Flexibility and Formulation
Chris Marble	Environmental Horticulture	Mid-Florida REC	Developing Postemergence Weed Control Strategies for Nonturf Groundcovers in Florida
Sandra Wilson	Environmental Horticulture	Gainesville Campus	Introduction of New Native Plants to Florida's Green Industry
Carlee Steppe	Environmental Horticulture	Gainesville Campus	

2019-2020

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Andrew K. Koeser	Environmental Horticulture	Gulf Coast REC	Tree Preservation Ordinances in the State of Florida – How does Policy Impact Canopy Coverage?
Deborah R. Hilbert	Environmental Horticulture	Gulf Coast REC	
Drew C. McLean	Environmental Horticulture	Gulf Coast REC	
Alexander J. Reisinger	Soil and Water Sciences	Gainesville Campus	Quantifying Nitrogen Leaching from Residential Soils in Florida
Eban Bean	Agricultural and Biological Engineering	Gainesville Campus	
Mark Clark	Soil and Water Sciences	Gainesville Campus	
Tom Yeager	Environmental Horticulture	Gainesville Campus	Automated Rain Gauge Device to Monitor Container Drainage for Irrigation Management
Jeff Million	Environmental Horticulture	Gainesville Campus	
Laura Warner	Agricultural Education and Communication	Gainesville Campus	Environmentally Friendly Landscaping: Addressing a Need for the Communications Research
Michael Dukes	Agricultural and Biological Engineering	Gainesville Campus	
Esen Momol	Center for Landscape Conservation & Ecology	Gainesville Campus	
Eban Bean	Agricultural and Biological Engineering	Gainesville Campus	Optimizing Soil Amendment Characteristics for Improving Environmental and Resource Sustainability
Michael Dukes	Agricultural and Biological Engineering	Gainesville Campus	
Wagner Vendrame	Environmental Horticulture	Tropical REC	Pilot Study on Management Strategies of Hibiscus Bud Weevil
Catharine Mannion	Entomology and Nematology	Tropical REC	
Romina Gazis	Plant Pathology	Tropical REC	
Adam G. Dale	Entomology and Nematology	Gainesville Campus	Determining the Effects of St. Augustinegrass Cultivar Diversity on Belowground Ecosystem Processes
Dorota Porazinska	Entomology and Nematology	Gainesville Campus	
Xavier Martini	Entomology and Nematology	North Florida REC	Survey of the Invasive Mite *Phyllocoptes Fructiphilus* Rose Rosette Virus (RRV) and of its Predatory Mites in Northern Florida
Austin N. Fife	Entomology and Nematology	North Florida REC	
Catharine Mannion	Entomology and Nematology	Tropical REC	Hibiscus Bud Weevil – A New Threat to Hibiscus Production
William Schall	IFAS Extension	Tropical REC	
Alfred Huo	Environmental Horticulture	Mid-Florida REC	Effect of Carbon and SiO2 Nanoparticles on Rooting and Growth of Different Ornamental Plants
Roger Kjelgren	Environmental Horticulture	Mid-Florida REC	

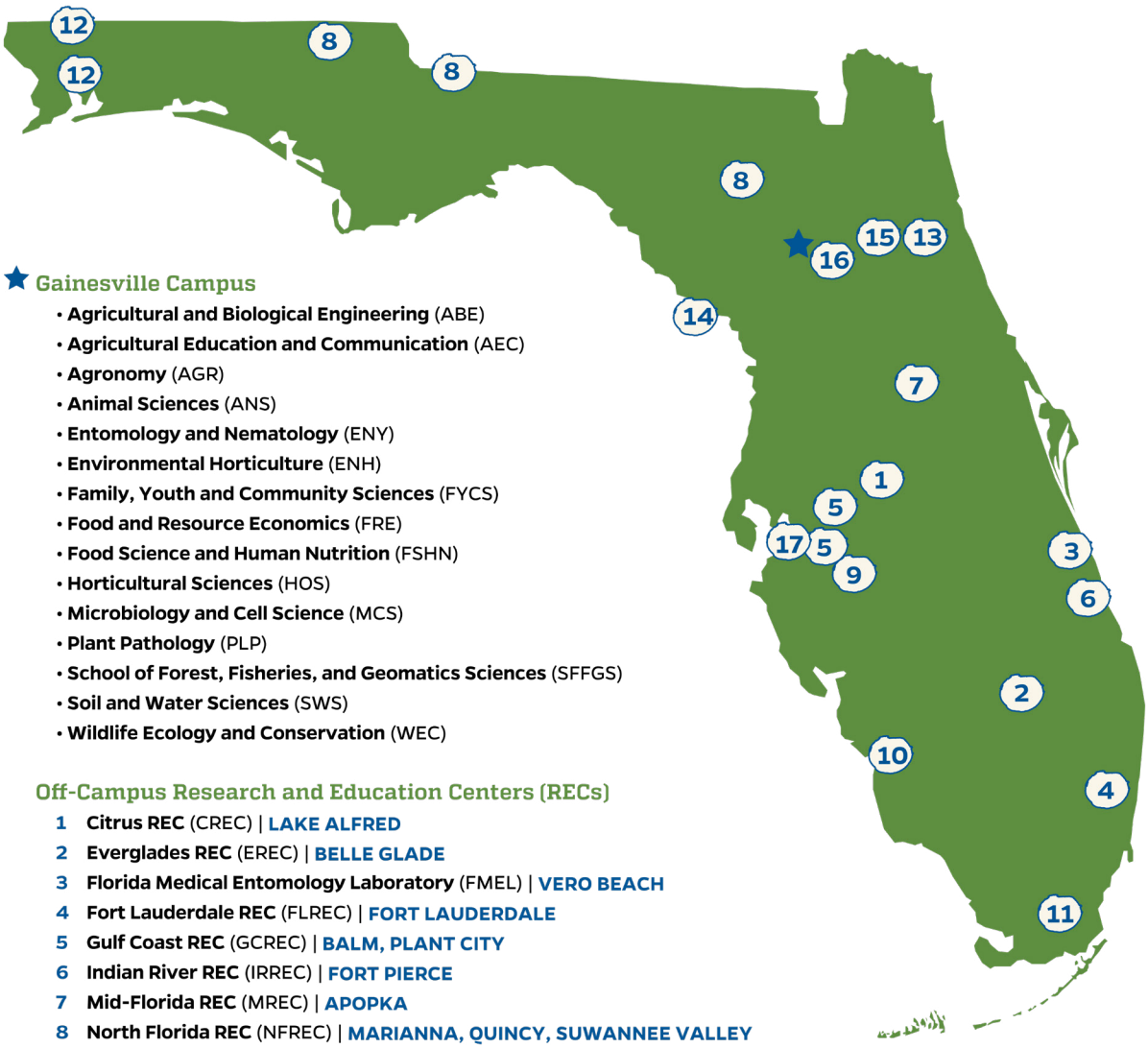
2020-2021

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Mysha Clarke	School of Forest, Fisheries, and Geomatics Sciences	Gainesville Campus	The role of gardening activities on resilience quality of life (especially during the COVID-19 pandemic)
Andrew Koeser	Environmental Horticulture	Gulf Coast REC	Determining Minimum Planting Widths for the Small-Stature Trees in Compact Developments
Deb Hilbert	Environmental Horticulture	Gulf Coast REC	
Drew McLean	Environmental Horticulture	Gulf Coast REC	
Marco Schiavon	Environmental Horticulture	Ft. Lauderdale REC	Construction of plots for long term evaluation of effects of effluent water on turfgrass
Bryan Unruh	Environmental Horticulture	West Florida REC	Establishment and Evaluation of Mixed Species Landscapes Comprising Perennial Grasses and Legumes
Ann Blount	Agronomy	North Florida REC	
Adam Dale	Entomology and Nematology	Gainesville Campus	
Thomas Yeager	Environmental Horticulture	Gainesville Campus	Reducing Nutrient Loss from Containers
Jeff Million	Environmental Horticulture	Gainesville Campus	
Brian Bahder	Entomology and Nematology	Ft. Lauderdale REC	Measuring degradation of insect and phytoplasma DNA on sticky traps
Adam Dale	Entomology and Nematology	Gainesville Campus	Developing methods for biodiversity-certified ornamental plant production
Jaret Daniels	Floridam Museum of Natural History	Gainesville Campus	
Chris Marble	Environmental Horticulture	Mid-Florida REC	Finding, Evaluating, and Fine-tuning Herbicide Alternatives to Glyphosate for the Florida Landscape Industry
Anthony Witcher	Tennessee State University		
Gary Vallad	Plant Pathology	Gulf Coast REC	Viburnum Foliar Disease Management; Downy Mildew & Cercospora Leaf Spot
Shawn Steed	Extension Agent III	Hillsborough City	
Fernando Alferez	Horticultural Sciences	Southwest Florida REC	Improving seed production and availability of major citrus rootstocks by determining seed viability during maturation and storage
Manjul Dutt	Horticultural Sciences	Citrus REC	

2021-2022

PI/Co-PI NAME	HOME UNIT	LOCATION	TITLE
Basil Iannone	Forest, Fisheries & Geomatics Sciences	Main Campus	How does plant diversity, vegetation structure, and management contribute to ecosystem services in residential landscaping?
Jesse Jones	Forest, Fisheries & Geomatics Sciences	Main Campus	
Brian Bahder	Entomology and Nematology	Ft. Lauderdale REC	Assessment of insecticides for control of Haplaxius crudus, the vector of lethal bronzing
De-Fen Mou	Entomology and Nematology	Ft. Lauderdale REC	
Braham Dhillon	Plant Pathology	Ft. Lauderdale REC	Detecting overlap of pathogen presence and trunk rot in palms
Adam Dale	Entomology & Nematology	Main Campus	Integrating Pest and Pollinator Management Strategies for Ornamental Plant Production
Jaret Daniels	Florida Museum of Natural History	Main Campus	
Bernadette Mach	Entomology & Nematology	Main Campus	
Ramdas Kanissery	Horticultural Sciences	Southwest Florida REC	"Place it and forget it" - Super absorbent medium for long-term weed suppression and plant-safe herbicide placement in nursery production
Stephen "Chris" Marble	Environmental Horticulture	Mid-Florida REC	Finding, Evaluating, and Fine-tuning Herbicide Alternatives to Glyphosate for the Florida Landscape Industry: PART II
Gary Vallad	Plant Pathology	Gulf Coast REC	Viburnum Foliar Disease Management: Disease mitigation during plant propagation
Shawn Steed	Environmental Horticulture	UF/IFAS Extension	
Wael Elwakil	Hillsborough County Extension	UF/IFAS Extension	
Wagner Vendrame	Environmental Horticulture	Main Campus	Improved Foliage Production Using Micropropagation - The Monstera Model
Jianping Wang	Agronomy	Main Campus	Developing a rapid molecular method to assess sugarcane mosaic virus (SCMV) load in turfgrass breeding materials
Kevin Kenworthy	Agronomy	Main Campus	
Philip Harmon	Plant Pathology	Main Campus	

UF/IFAS Research Units



★ **Gainesville Campus**

- **Agricultural and Biological Engineering** (ABE)
- **Agricultural Education and Communication** (AEC)
- **Agronomy** (AGR)
- **Animal Sciences** (ANS)
- **Entomology and Nematology** (ENY)
- **Environmental Horticulture** (ENH)
- **Family, Youth and Community Sciences** (FYCS)
- **Food and Resource Economics** (FRE)
- **Food Science and Human Nutrition** (FSHN)
- **Horticultural Sciences** (HOS)
- **Microbiology and Cell Science** (MCS)
- **Plant Pathology** (PLP)
- **School of Forest, Fisheries, and Geomatics Sciences** (SFFGS)
- **Soil and Water Sciences** (SWS)
- **Wildlife Ecology and Conservation** (WEC)

Off-Campus Research and Education Centers (RECs)

1 **Citrus REC** (CREC) | **LAKE ALFRED**
2 **Everglades REC** (EREC) | **BELLE GLADE**
3 **Florida Medical Entomology Laboratory** (FMEL) | **VERO BEACH**
4 **Fort Lauderdale REC** (FLREC) | **FORT LAUDERDALE**
5 **Gulf Coast REC** (GCREC) | **BALM, PLANT CITY**
6 **Indian River REC** (IRREC) | **FORT PIERCE**
7 **Mid-Florida REC** (MREC) | **APOPKA**
8 **North Florida REC** (NFREC) | **MARIANNA, QUINCY, SUWANNEE VALLEY**
9 **Range Cattle REC** (RCREC) | **ONA**
10 **Southwest Florida REC** (SWFREC) | **IMMOKALEE**
11 **Tropical REC** (TREC) | **HOMESTEAD**
12 **West Florida REC** (WFREC) | **JAY, MILTON**

Research and Demonstration Sites

13 **Hastings Agricultural Extension Center** (HAEC) | **HASTINGS**
14 **Nature Coast Biological Station** (NCBS) | **CEDAR KEY**
15 **Ordway-Swisher Biological Station** (OSBS) | **MELROSE**
16 **Plant Science Research and Education Unit** (PSREU) | **CITRA**
17 **Tropical Aquaculture Laboratory** (TAL) | **RUSKIN, APOLLO BEACH**

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