CLEMSON® UNIVERSITY

Plant and Pest Diagnostic Clinic
Molecular Pathogen and Pest Detection Lab
Commercial Turfgrass Clinic
Nematode Assay Lab

2023 ANNUAL REPORT

The cover page image highlights urediniospores of *Pseudocerradoa paullula*, a new pathogen detected in South Carolina, USA in 2023. See **Appendix I** for details.

The information contained in this report is based upon work that was partly supported by the National Plant Diagnostic Network (NPDN). NPDN is a consortium of plant diagnostic labs across all 50 U.S. states and several territories supported by the U.S. Department of Agriculture, National Institute of Food and Agriculture.

A message from Plant Industry Labs staff

As a part of Clemson University's Regulatory Services, The Department of Plant Industry laboratories (DPI Labs) is a unique program serving stakeholders in South Carolina and clients in other states by providing timely and high-quality diagnostic services such as plant healthy analysis, pest ID, plant/weed ID, nematode assay, fungicide-resistance profiling, and molecular identifications and rapid detection services. DPI Labs includes Plant and Pest Diagnostic Clinic (PPDC), the state lab dedicated to handle all plant diagnostic samples in South Carolina, the Molecular Pathogen and Pest Detection (MPPD) Laboratory, the lab providing a variety of specialty services using advanced molecular techniques, and the Commercial Turfgrass Clinic (CTC), a specialty lab providing expedited, high-quality diagnostic services of turf problems to golf courses, athletic fields, sod farms, and other turf management professionals. The Nematode Assay Lab (NAL) at the Clemson University Department of Plant and Environmental Sciences (PES) serves under contractual agreement with PPDC to identify plant parasitic nematodes. The 2023 Annual Report of NAL is included as **Appendix II**. DPI Labs also partners with other programs at the Clemson Regulatory Services, as well as Clemson Extension, teaching, and research communities to document new diseases and pests in South Carolina and provide educational, research, and training opportunities.

Dr. Jacob Taylor joined PPDC in November 2023 to begin his new position as a Plant Taxonomist. He has been leading the diagnostic services including plant/weed ID and molecular identifications of mushrooms. Jacob has also been working on developing collaborations with the DPI Invasive Species Program, Clemson Extension, and other Clemson departments, aiming at enhancing the detection and management of noxious weeds.

Jordan Withycombe joined PPDC in March 2024 as a Lab Technologist II. In her role, she is primarily responsible for performing plant diagnoses. She has also been assisting the Manager and the Coordinator in laboratory management and coordination tasks, such as outreach, intern hiring and training, and sample receiving, entry, and coordination.

Ray Adcock has been working as a Designer for the NPDN Communicator Newsletters since September 2023. He is an Info. Sys. Business Analyst II at Clemson Public Service and Agriculture (PSA).

We thank faculty members, specialists, agents, and retired professionals who provided diagnostic and professional consultation in 2023. The Annual DPI Labs Advisory Committee Meeting took place on December 13, 2023. A special thanks to our Advisory Committee members who attended the meeting and provided suggestions and ideas that are paramount for improving our services and program. We also thank our dedicated student interns in 2023.

The DPI Labs provided nine lab tours in 2023. The attendees included 64 students and six staff and five faculty members. We welcome old and new friends to come tour the DPI Labs in 2024!

DPI Labs Staff Members

- Predeesh Chandran (Entomologist II)
- Dr. Curt Colburn (Molecular Biologist and Turf Diagnostician)
- Diana Low (Coordinator)
- Dr. Jacob Taylor (Plant Taxonomist and Weed Scientist)
- Jordan Withycombe (Laboratory Technologist II and Plant Diagnostician)
- Dr. Xiao Yang (Manager)

Graduate Diagnostic Assistants

- Annie Bruno
- Atiya Khan
- Daniela Negrete Moreno

Graduate Research Assistant

- Amanda Minner

Undergraduate Diagnostic Assistant

- Charles McGuinn

Advisory Committee Members

- Dr. Shannon Alford (Director, Agricultural Service Laboratory)
- Dr. Elizabeth (Libby) Cieniewicz (Assistant Professor of Plant Virology, PES)
- Dr. Timothy Drake, Jr. (State Entomologist, Regulatory Services Programs)
- Matt Howle (Manager and State Survey Coordinator, Invasive Species Program, DPI)
- Dr. Anthony Keinath (Professor, Vegetable Pathologist, PES)
- Dr. Julia Kerrigan (Associate Professor of Mycology, PES)
- Dr. Haiying Liang (Associate Professor, Department of Genetics and Biochemistry)
- Steven Long (Assistant Director, State Plant Regulatory Official, DPI)
- Dr. Joseph Roberts (Associate Professor of Turfgrass Pathology and Nematology, PES)
- Dr. Guido Schnabel (Professor of Plant Pathology – Fruit Crops, PES)
- Dr. Hehe Wang (Assistant Professor of Plant Bacteriology, PES)
- Dr. Ted Whitwell (Professor Emeritus)
- Meg Williamson (Educational Plant Diagnostician, DPI)

Other Advisory Consultants in 2023

- Dr. Lance Beecher (Aquaponics, Aquaculture and Fisheries Specialist, Clemson Extension)
- Dr. Eric Benson (Professor, Extension Entomologist, PES)
- Dr. Tom Bilbo (Assistant Professor of Vegetable & Strawberry Entomology, PES)
- Dr. Juang Chong (Adjunct Professor of Entomology)
- Alex Coleman (Extension Small Grains & Sorghum Specialist)

- Millie Davenport (Director, Home and Garden Information Center)
- Dr. Lorena Endara (Herbarium Curator, Lecturer, Department of Biological Sciences)
- Dr. John Hains (Associate Professor Emeritus)
- Dr. W. Cory Heaton (Assistant Director, Extension Wildlife Specialist, Clemson Extension)
- Dr. Steven Jeffers (Professor of Plant Pathology – Ornamental Crops & Trees, PES)
- Dr. Churamani Khanal (Assistant Professor of Plant Nematology, PES)
- Lambert (Bert) McCarty (Professor of Turfgrass Science & Management, PES)
- S. Cory Tanner (Horticulture Program Team Director, PES)
- Dr. Joey Williamson (Retired Clemson Extension Horticulture Agent)

Table of Contents

Plant and Pest Diagnostic Clinic (PPDC) Report

A total of 1207 samples were received and processed at PPDC in 2023, an increase of approximately 19% compared to 2022 (**Fig. 1**). Summer remained as our busiest season with more than 107 samples received in each month between April and October (**Fig. 2**).

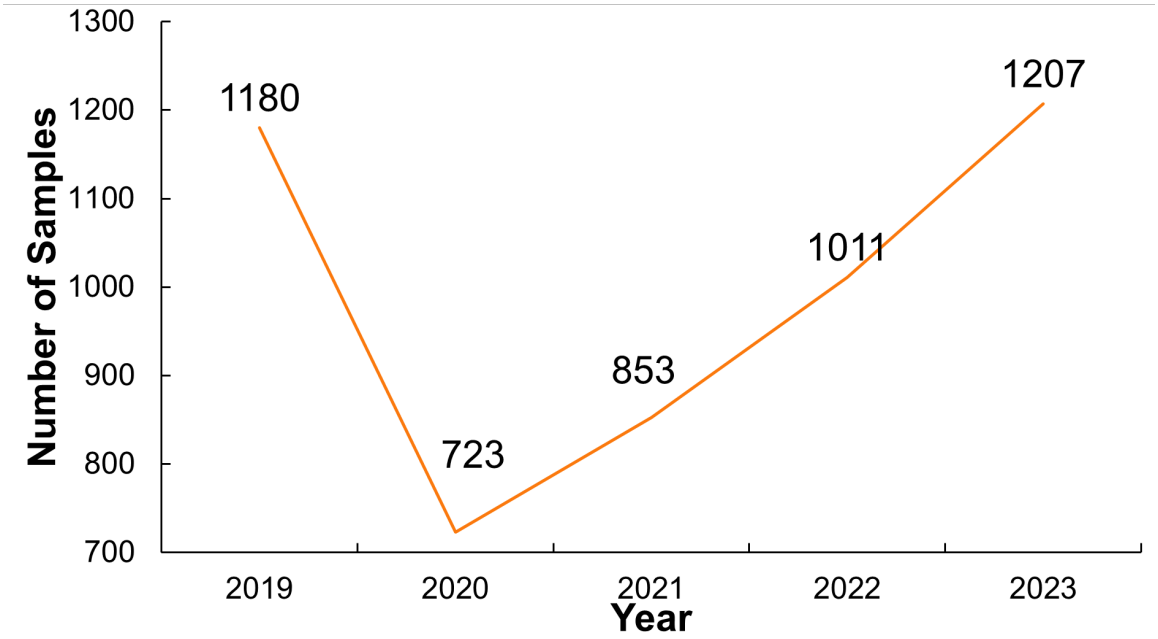

Fig. 1 Number of samples processed at PPDC by year over the past five years

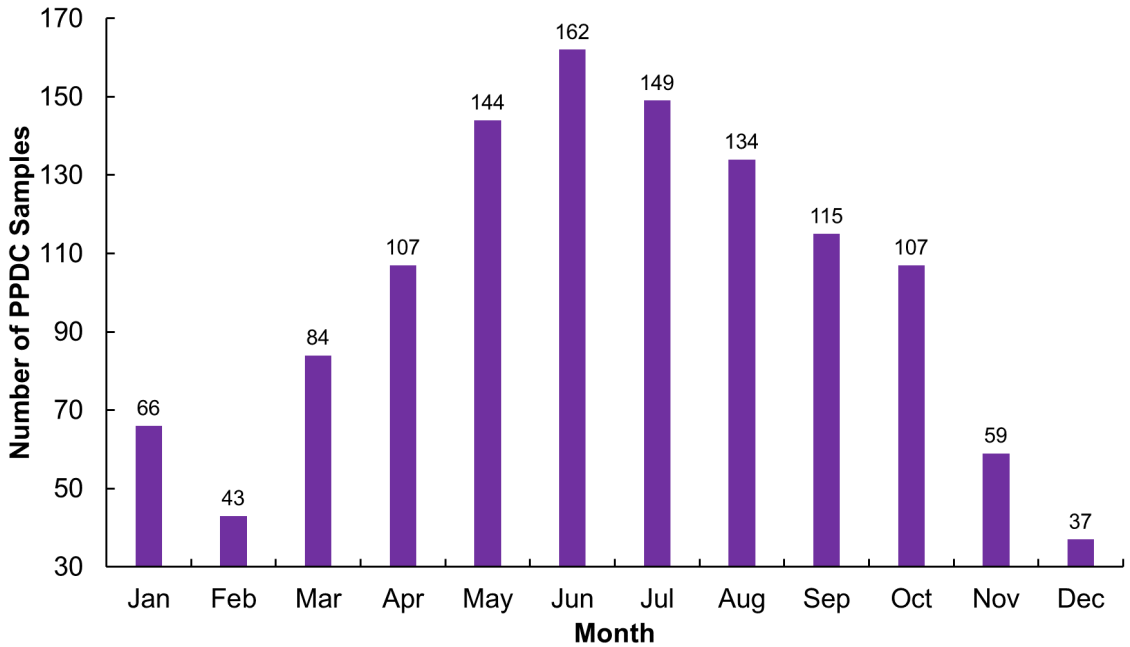

Fig. 2 Number of samples processed at PPDC by month in 2023

A total of 752 PPDC samples in 2023 were received from county extension offices. A total of 635 samples originated from residential sites, while 569 samples were submitted by commercial clients (**Fig. 3**). We provided services to a total of 1805 clients in 2023. Most of our clients (approximately 60%) were homeowners, home gardeners, and other residential clients referred by extension agents. (**Fig. 4**).

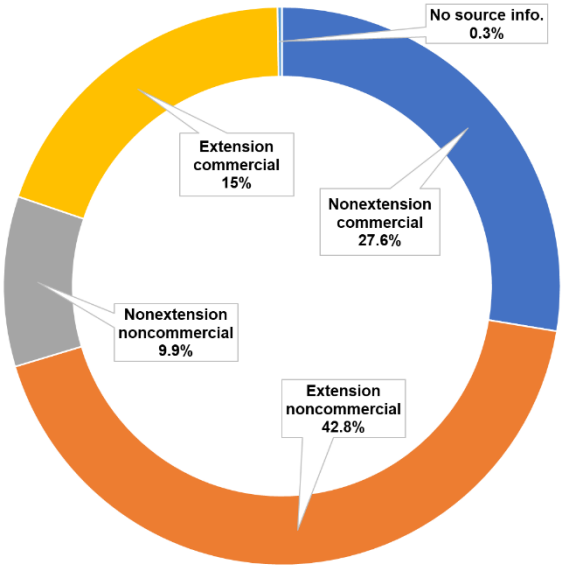

Fig. 3 Percentage of samples by source at PPDC in 2023

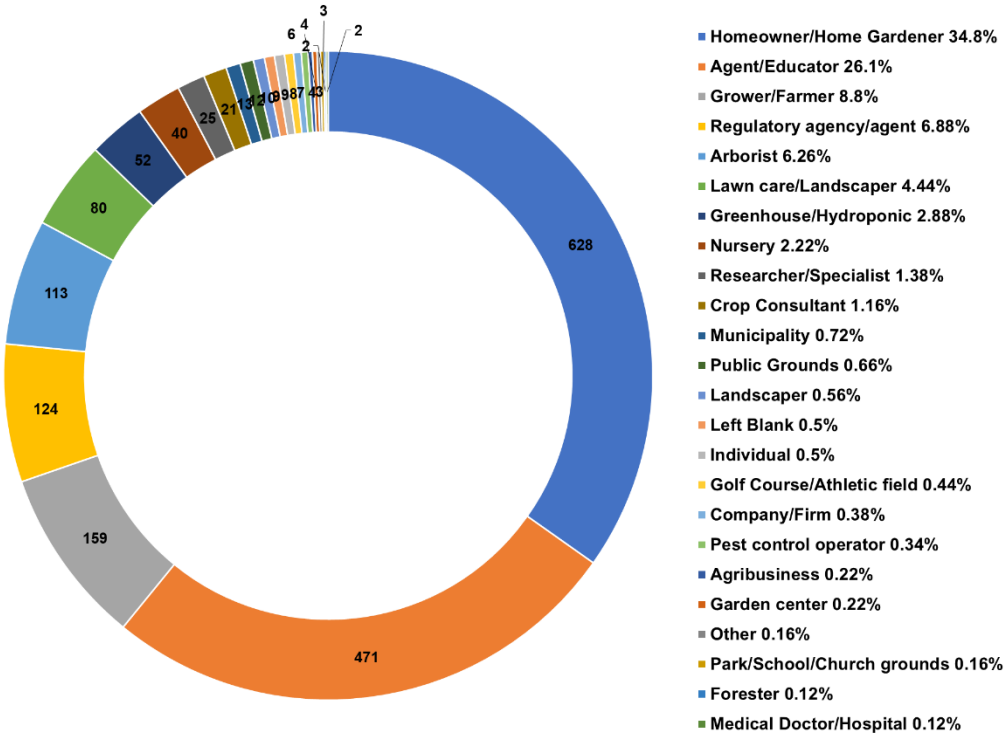

Fig. 4 PPDC sample counts by client type in 2023

PPDC samples in 2023 belonged to at least 16 categories. More than 42% of samples were woody ornamental plants. We found an increase in submissions of samples seeking insect ID and plant health analyses for small fruits (**Fig. 5**).

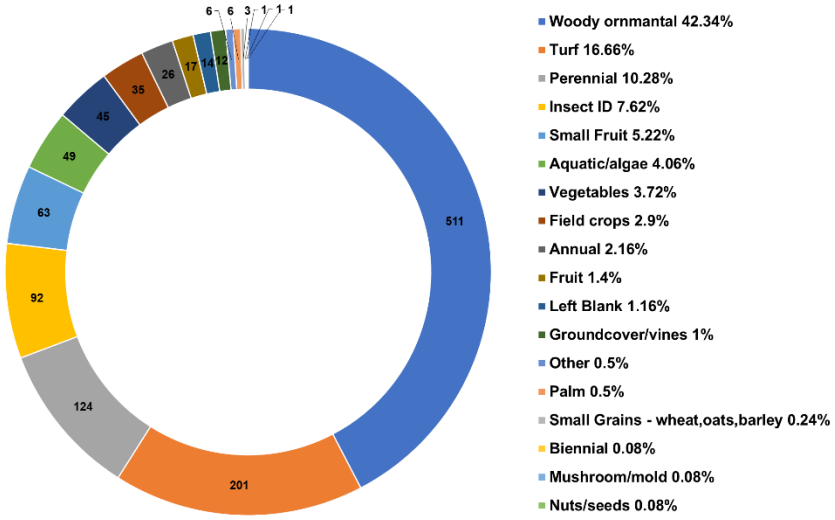

Fig. 5 PPDC sample counts by sample category in 2023

A total of 1132 samples in 2023 came from 44 SC counties (**Fig. 6**). Other clients were from U.S. states including AL (2), CA (5), FL (4), GA (4), IL (1), KS (12), ME (1), MI (1), MO (2), MS (3), NC (8), NY (1), OR (8), PA (2), TN (7), TX (5), VA (5), and WA (4).

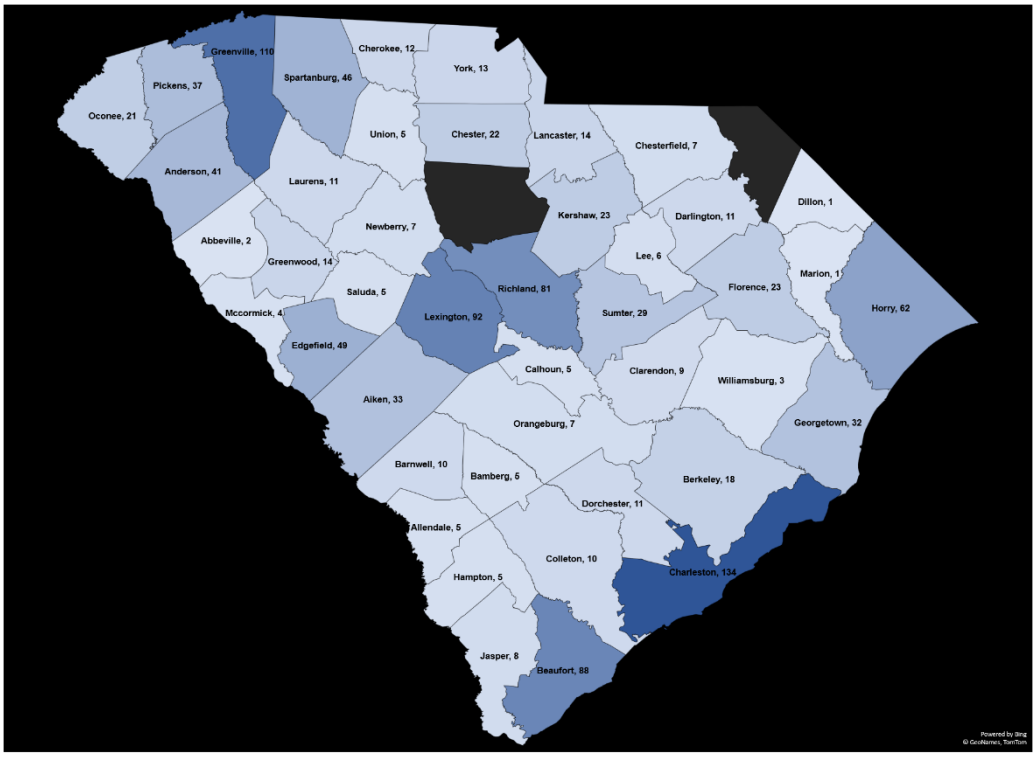

Fig. 6 Count of PPDC samples in 2023 by county in SC

Molecular Pathogen and Pest Detection (MPPD) Lab Report

The MPPD Lab utilizes polymerase chain reaction (PCR)-based assays to detect plant pathogens and pests (e.g., Africanized honeybees and two honeybee pathogens, the American foulbrood pathogen *Paenibacillus larvae* and the European foulbrood pathogen *Melissococcus plutonius*). The main targets are plant diseases and pathogens that are of regulatory concern by USDA APHIS and the state of South Carolina, such as the sudden oak death pathogen *Phytophthora ramorum* and the citrus greening pathogens '*Candidatus* Liberibacter africanus', '*C*. Liberibacter asiaticus', and '*C*. Liberibacter americanus'. The MPPD Lab also provides fungicide-resistance testing services for *Botrytis* and *Colletotrichum*. We also work closely with other Plant Industry programs to conduct state-wide surveys such as the Cooperative Agricultural Pest Survey (CAPS).

A total of 118 samples were processed at the MPPD Lab in 2023. Most samples were of regulatory concern. Sample counts by category are listed in **Table 1**.

Table 1 Sample count by category at the MPPD Lab in 2023

Sample Category	Number of Samples
Fungicide-resistance Testing	27
Microbial Testing	23
Fungus ID by DNA Sequencing	20
Other	20
Phytophthora ramorum qPCR Rapid Detection	13
Neopestalotiopsis Rapid Detection by PCR-RFLP	9
Honey Bee Testing (Africanized Honey Bee and Foulbroods)	6

Commercial Turfgrass Clinic (CTC) Report

A total of 39 samples were processed at CTC, which had a slight increase on top of the 2022 sample load (34 samples). These included three bentgrass, 32 bermudagrass, and four zoysia grass samples. Diagnostic results at CTC in 2023 are listed in **Table 2**.

Table 2 Diagnostic results at CTC in 2023

Host Name	Pest Name	Confirmed	Suspected
Bentgrass (*Agrostis* sp./spp.)	Root-knot nematodes (*Meloidogyne* sp./spp.)	0	2
	Pythium root dysfunction (*Pythium* sp./spp.)	1	0
Bermudagrass (*Cynodon* sp./spp.)	Brown patch (*Rhizoctonia solani*)	5	0
	Free living nematodes (Multiple genera)	0	1
	Pythium root dysfunction (*Pythium* sp./spp.)	4	1
	Take-all (*Gaeumannomyces* sp./spp.)	8	0
	Sting nematodes (*Belonolaimus* sp./spp.)	0	1
	Anthracnose (*Colletotrichum* sp./spp.)	2	0
	Curvularia blight; Leaf spot (*Curvularia* sp./spp.)	4	0
	Root-knot nematodes (*Meloidogyne* sp./spp.)	5	0
	Cereal/ grass disease (*Bipolaris* sp./spp.)	1	0
	Large patch (*Rhizoctonia solani*)	2	0
	Pythium blight; Cottony blight (*Pythium* sp./spp.)	1	0
Zoysia Grass (*Zoysia* sp./spp.)	Large patch (*Rhizoctonia solani*)	1	0
	Curvularia blight; Leaf spot (*Curvularia* sp./spp.)	1	0
	Take-all (*Gaeumannomyces* sp./spp.)	2	0

Appendix I: First report of *Pseudocerradoa paullula* causing aroid leaf rust on Swiss cheese plant *Monstera deliciosa* in South Carolina, USA

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In February 2023, two *Monstera deliciosa* Liebm. (Araceae) plants with typical symptoms of leaf rust disease were detected at a grocery store in Oconee Co., South Carolina. Symptoms included chlorotic leaf spots and abundant brownish uredinia, mainly on the adaxial surface of more than 50% of leaves. The same disease was detected on 11 out of 481 *M. deliciosa* plants in a greenhouse at a plant nursery located in York Co., South Carolina, in March 2023. The first plant sample detected in February was used for morphological characterization, molecular identification, and pathogenicity confirmation of the rust fungus. Urediniospores were densely aggregated, globose, golden to golden brown in color, and measured 22.9 to 27.9 µm (aver. 26.0 ± 1.1 µm; *n*=50) in diameter with wall thickness at 1.3 to 2.6 µm (aver. 1.8 ± 0.3 µm; *n*=50). Telia were not observed. These morphological traits aligned with those of *Pseudocerradoa paullula* (basionym: *Puccinia paullula*; Ebinghaus et al. 2022; Sakamoto et al. 2023; Sydow and Sydow 1913; Urbina et al. 2023). Genomic DNA was extracted from urediniospores collected from the naturally infected plant sample and used for PCR amplification and DNA sequencing of the large subunit (LSU) genetic marker with primers LRust1R and LR3 (Vilgalys and Hester 1990; Beenken et al. 2012). The LSU sequence of the rust fungus in South Carolina (GenBank accession: OQ746460) is 99.9% identical to that of *Ps. paullula* voucher BPI 893085 (763/764 nt.; KY764151), 99.4% identical to that of voucher PIGH 17154 in Florida, USA (760/765 nt.; OQ275201), and 99% identical to that of voucher TNS-F-82075 in Japan (715/722 nt.; OK509071). Based on its morphological and molecular characteristics, the causal agent was identified as *Ps. paullula*. This pathogen identification was also corroborated by the U.S. Department of Agriculture, Animal and Plant Health Inspection Service, Plant

Pathogen Confirmatory Diagnostics Laboratory in Laurel, Maryland. To confirm the fungus's pathogenicity on *M. deliciosa* and *M. adansonii* Schott (Sakamoto et al. 2023), three plants of each *Monstera* species were inoculated by spraying with a suspension of urediniospores collected from the original plant sample (1×10^6 spores per ml; approx. 40 ml per plant). Three non-inoculated control plants of each host species were treated with deionized water in the same manner. Plants were placed in a plastic tray with wet paper towels to maintain moisture. The tray was placed at 22°C for an 8-h photoperiod and covered for five days to facilitate infection. On 25 days after inoculation, abundant spots bearing urediniospores were produced on all leaves of inoculated *M. deliciosa* plants. A few uredinia were observed on two of the three inoculated *M. adansonii* plants. All non-inoculated control plants remained asymptomatic. Morphological features of urediniospores collected from inoculated plants matched those of *Ps. paullula* used as the inoculum. Aroid leaf rust on *Monstera* plants was officially reported in Australia, China, Japan, Malaysia, Philippines, and Florida, USA (Shaw 1991; Sakamoto et al. 2023; Urbina et al. 2023). This is the first report of *Ps. paullula* causing this disease on *M. deliciosa* in South Carolina, USA. *Monstera* species are popular indoor and landscape plants. Potential impact and regulatory responses regarding *Ps. paullula*, a newly introduced and rapidly spreading pathogen in the USA, warrant further evaluation and discussion.

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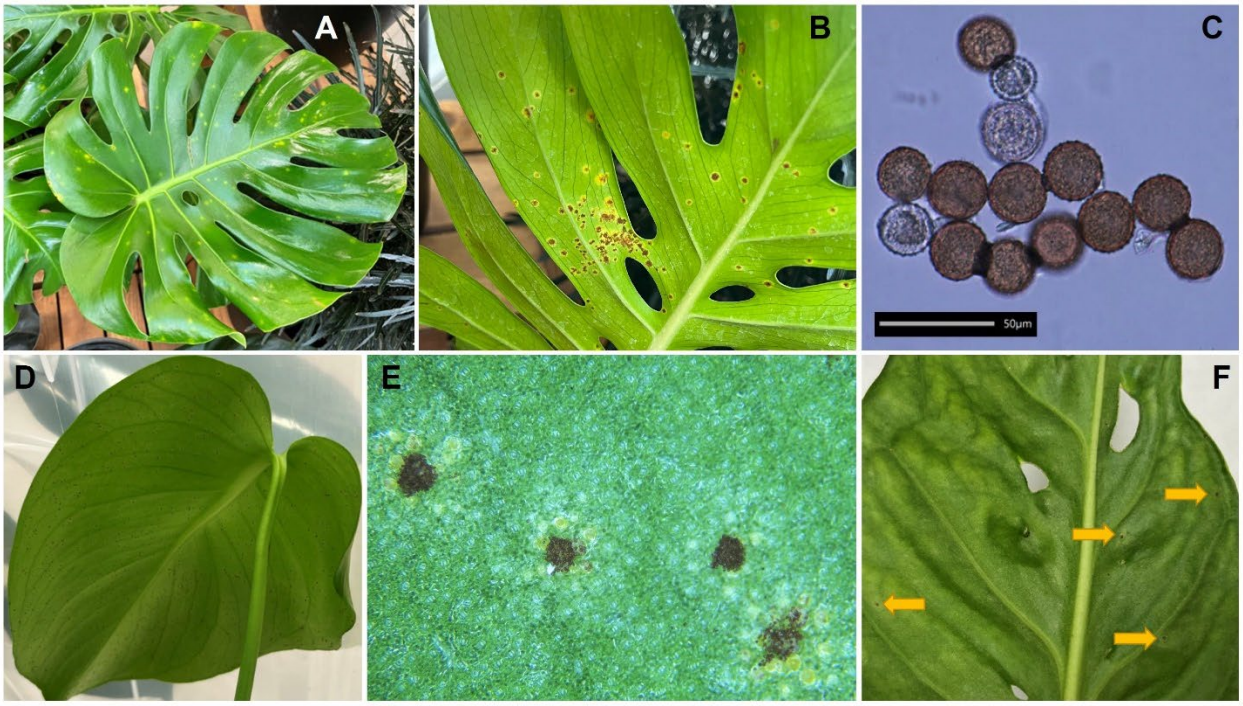

Fig. A1 (A to C) Aroid leaf rust caused by *Pseudocerradoa paullula* on the naturally infected *Monstera deliciosa* sample detected at a grocery store in South Carolina, USA. (A) Chlorotic spots on the abaxial leaf surface. (B) Spots bearing uredinia on the adaxial leaf surface. (C) Globose, golden to golden brownish urediniospores. (D to F) Symptoms of aroid leaf rust disease on artificially inoculated plants 25 days after inoculation. (D) Uredinia produced on an inoculated *Monstera deliciosa* leaf. (E) Four uredinia with aggregated urediniospores on the adaxial leaf surface of *M. deliciosa*. (F) A few uredinia (arrows) produced on an inoculated *M. adansonii* leaf.

Appendix II: Nematode Assay Lab 2023 Annual Report

The Nematode Assay Lab (NAL) is led by Dr. Churamani Khanal. It locates in the Biosystems Research Complex at the Department of Plant and Environmental Sciences of Clemson University. NAL serves under contractual agreement with PPDC to extract and identify plant parasitic nematodes from plant and soil samples and to provide management recommendations, as needed. The nematode assay samples are submitted from various sources, including extension offices, commercial operations, research projects, and regulatory inspectors. In 2023, NAL processed 911 samples including over 130 samples received in each of March and April (**Fig. A2**).

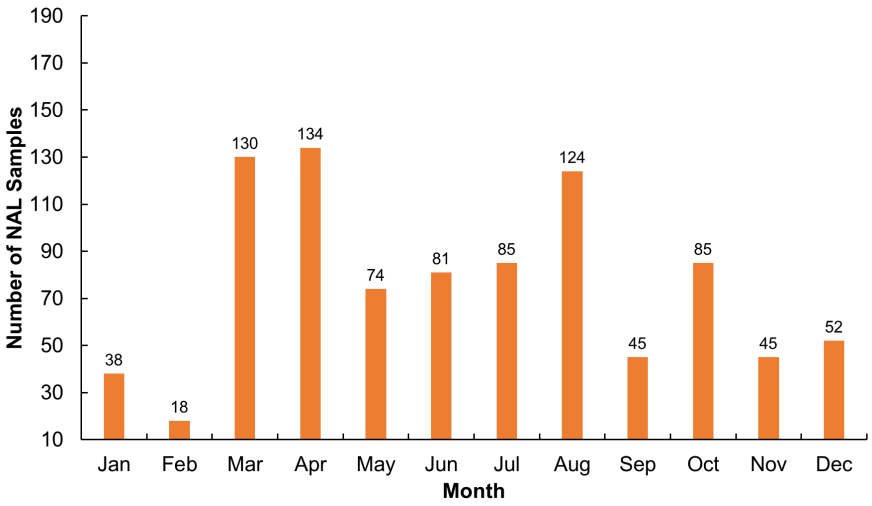

Fig. A2 Number of samples processed at NAL by month in 2023

Most samples received at NAL in 2023 (approx. 95%) were from commercial sources (**Fig. A3**). NAL provided services to 1760 clients in 2023.

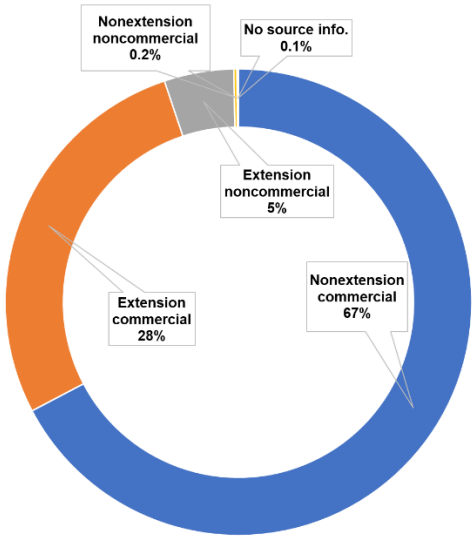

Fig. A3 Count and Percentage of samples by source at NAL in 2023

More than 46% of the NAL samples originated from turfgrasses. A total of 619 samples came from 41 counties in SC (**Fig. A4**). Out-of-state clients of NAL in 2023 were AR (2), AZ (1), FL (148), GA (7), LA (3), NC (34), NV (58), NY (34), TN (2), and TX (2), plus one sample with unknown state origin.

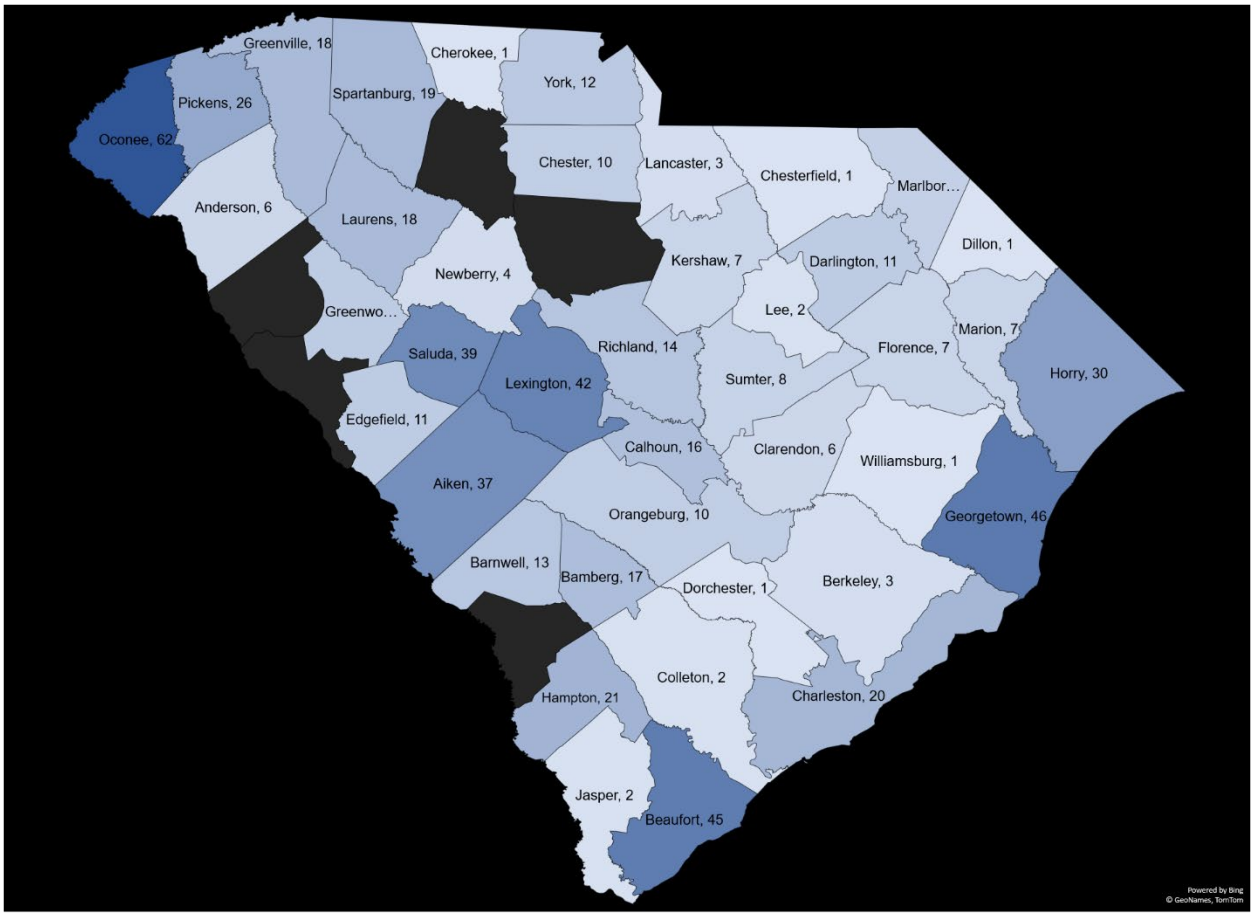

Fig. A4 Count of NAL samples by SC county in 2023