

#### TENNESSEE STATE UNIVERSITY College of Agriculture

# **October 1-2, 2024** McMinnville, TN



# VASCULAR STREAK DIEBACK Meeting proceedings

**Planning a Solution:** A Partnership to Identify Research and Extension Priorities for Vascular Streak Dieback (VSD) in Woody Ornamental Crops

## Sponsored, Hosted, & Published By:

USDA-NIFA Specialty Crops Research Initiative (Award No. 2024-51181-43200)



# Vascular Streak Dieback Meeting Proceedings

Planning a Solution: A Partnership to Identify Research and Extension Priorities for Vascular Streak Dieback (VSD) in Woody Ornamental Crops

Tennessee State University, Otis L. Floyd Nursery Research Center, McMinnville, TN October 1-2, 2024

> Sponsored by USDA-NIFA Specialty Crops Research Initiative (Award No. 2024-51181-43200)

> *Meeting hosted by* **Tennessee State University College of Agriculture**

**Published by** Horticultural Research Institute, The AmericanHort Foundation



ENNESSEE USDA National Institute of Food and Agriculture



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#### Acknowledgements

Many people contributed to the success of the 2024 Vascular Streak Dieback Meeting, from the Tennessee State University College of Agriculture team who helped plan and support it to everyone who presented at and attended the meeting. We thank them all for their excellent support, contributions, and involvement.

We especially acknowledge the support of Dr. Thomas A. Bewick (USDA Specialty Crop Research Initiative Program Leader) and the USDA-NIFA Specialty Crop Research Initiative (SCRI) Planning Grant (**Award No. 2024-51181-43200**) titled "*Planning a Solution: A Partnership to Identify Research and Extension Priorities for Vascular Streak Dieback (VSD) in Woody Ornamental Crops.*" The grant enabled the presenters and specialty crop industry stakeholders to come together in support of the meeting's main objective: "To develop a comprehensive research and extension plan for nursery crops that addresses stakeholder needs for improved, cost-effective, and sustainable vascular streak dieback management." The meeting represented a vitally important step in the efforts to address the growing VSD challenge, and it would have been impossible without the direct support and facilitation of USDA-NIFA-SCRI.

Special thanks to Dr. Karla Addesso (Director of TSU's Otis L. Floyd Nursery Research Center, or NRC), who made it possible to host the meeting at the TSU-NRC, and who assisted directly with many of the meeting's logistical aspects, as did Holly Hodges (TSU-NRC Financial Analyst). We also offer a big thanks to the other members of the committee and support team highlighted below for all they did to help make the meeting a success.

Finally, we would like to express our sincere gratitude to the Horticultural Research Institute, the AmericanHort Foundation, for its invaluable support and contribution in publishing the proceedings, and for its ongoing commitment to supporting scientific research for the advancement of the horticultural industry.

#### **Tennessee State University Local Arrangements Committee**

Dr. Fulya Baysal-Gurel, Chair Dr. Cansu Oksel Dr. Sujan Dawadi Terri Simmons Dr. Karla Addesso Dr. Kumuditha Hikkaduwa Epa Liyanage Holly Hodges

#### **Tennessee State University Local Arrangements Support**

Dr. Seyed Mohammad Rouhani	Pratima Subedi
Christina Jennings	Shahla Borzouei

### **Meeting Objective and Goals**

# Main Objective:

To develop a comprehensive research and extension plan for nursery crops that addresses stakeholder needs for improved, cost-effective, and sustainable vascular streak dieback management.

#### **Goals:**

- 1) Identify priorities and critical needs;
- 2) Identify knowledge gaps; and
- 3) Determine how to address priorities, critical needs, and knowledge gaps.

Otis L. Floyd Nursery Research Center Tennessee State University 472 Cadillac Lane, McMinnville, TN 37110 (931) 815-5155

USDA-NIFA Specialty Crops Research Initiative Planning a Solution: A Partnership to Identify Research and Extension Priorities for Vascular Streak Dieback (VSD) in Woody Ornamental Crops (Award No. 2024-51181-43200)

> October 1-2, 2024 All times are Central Standard Time Zone (CST) Zoom: <u>https://us02web.zoom.us/j/86203265530</u> Meeting ID: 862 0326 5530

#### Tuesday, October 1, 2024

8:00 – 8:30 AM	Arrival/registration and welcome	
8:30 – 8:50 AM	Opening statement (goals & objectives) and introductions	Dr. Fulya Baysal-Gurel (Tennessee State University)
8:50 – 9:30 AM	Understanding the problem: What is VSD? Distribution, host range, possible causal agent(s); Virginia survey results (30 min presentation; 10 min discussion)	Dr. John Bonkowski (Purdue University) Devin Bily (Virginia Dept. of Agriculture and Consumer Services)
9:30 – 9:50 AM	Multi-state survey results and national needs assessment survey results (10 min presentation; 10 min discussion)	Dr. Prabha Liyanapathiranage (Tennessee Dept. Agriculture)
9:50 – 10:00 AM	Break	
10:00 – 10:20 AM	Economic impact of VSD (10 min presentation; 10 min discussion)	Dr. Kumuditha Liyanage (Tennessee State University)
10:20 – 11:00 AM	Detection and diagnosis (30 min presentation; 10 min discussion)	Dr. John Bonkowski (Purdue University) Dr. Farhat Avin (Tennessee State University)
11:00 – 12:00 PM	Table group discussion 1: Open discussion of morning topics and key research/extension gaps (Tables to include mix of participants, i.e., growers, re-wholesalers, diagnosticians,	Table leaders

	researchers, extension personnel. Each table to select a leader/presenter and notetaker)	
12:00 – 1:00 PM	Lunch (will be provided)	
1:00 – 1:20 PM	Koch's postulates and/or Hill's criteria: Progress on assessing the relationship between <i>Csp</i> and VSD (10 min presentation; 10 min discussion)	Dr. Prabha Liyanapathiranage (Tennessee Dept. Agriculture)
1:20 – 1:50 PM	Predicted biology and epidemiology: Environmental factors and seasonality of VSD symptom development and spread; preliminary results from TSU's spore trapping trials; potential for VSD forecasting system (20 min presentation; 10 min discussion)	Dr. Prabha Liyanapathiranage (Tennessee Dept. Agriculture) Dr. Farhat Avin (Tennessee State University)
1:50 – 2:50 PM	Current and novel management strategies and challenges: Best-management practices; sanitation practices; chemical/biological control; identifying and utilizing natural host resistance and future focus on developing resistance germplasm; production methods to reduce susceptibility (fertilization, irrigation, container type/color, shade vs. light, tissue culture, grafting); pruning (root and upper canopy)	Dr. Fulya Baysal-Gurel (Tennessee State University) Dr. Amy Fulcher (University of Tennessee) Dr. Hsuan Chen (North Carolina State Univ.) Dr. Anthony Witcher (Tennessee State University) Dr. Jacob Shreckhise (USDA-ARS)
2:50 – 3:10 PM	Outreach/education training needs: Bilingual training of non-English-speaking workers; developing image libraries of diagnostic features; communicating effectively with stakeholders and students; impact assessment/evaluation	Dr. Fulya Baysal-Gurel (Tennessee State University) Dr. Kaitlin Barrios (Tennessee State University) Dr. Lana Petrie (Tennessee State University) Dr. John Ricketts (Tennessee State University)
3:10 – 3:20 PM	Break	
3:20 – 4:00 PM	Grower panel: Producer perspectives; overcoming challenges; approach to the coming growing season	Growers

4:00 – 4:15 PM	Table group discussion 2	Table leaders
4:15 – 4:45 PM	Table group presentations	Table leaders
4:45 – 5:00 PM	Closing remarks	Dr. Fulya Baysal-Gurel (Tennessee State University)
Wednesday, Octobe	er 2, 2024	
8:00 – 8:30 AM	Opening comments & compilation of priorities across table groups	Dr. Fulya Baysal-Gurel (Tennessee State University)
8:30 – 9:30 AM	Discipline-specific discussions: (Diagnostic, pathogen genetics, biology& epidemiology, host genetics, economics, sociology/behavioral aspects, management strategies, agricultural engineering, risk assessment, outreach, science communications, impact assessments/evaluation)	Small breakout groups
9:30 – 10:15 AM	Discipline-specific presentations	Group leaders
10:15 – 10:30 AM	Break	
10:30 – 12:00 PM	Proposal planning: Prioritize objectives & sub- objectives/obtainable goals	Team members
	Research/extension timelines: Identify short, medium, long-term goals	
12:00 – 1:00 PM		
	Lunch (will be provided)	
1:00 – 2:00 PM	Lunch (will be provided) Identify additional experts/cooperators needed: university, industry, government	Team members
1:00 – 2:00 PM	Identify additional experts/cooperators	Team members
1:00 – 2:00 PM	Identify additional experts/cooperators needed: university, industry, government	Team members
1:00 – 2:00 PM 2:00 – 2:15 PM	Identify additional experts/cooperators needed: university, industry, government Identify advisory board members	Team members Dr. Fulya Baysal-Gurel (Tennessee State University)

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#### **Abstracts and Papers**

#### Vascular Streak Dieback Symptomology, Signs, and Detection

John Bonkowski<sup>1</sup>, Tom Creswell<sup>1</sup>, Mike Munster<sup>2</sup>, Prabha Liyanapathiranage<sup>3</sup>, Fulya Baysal-Gurel<sup>3</sup>, and Devin Bily<sup>4</sup>

<sup>1</sup>Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907
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<sup>4</sup>Virginia Department of Agriculture and Consumer Services, Richmond, VA 23219

Between 2019 and 2021, nursery-grown 'Autumn Blaze' maple (Acer x freemanii 'Autumn Blaze') and redbuds (Cercis canadensis) showing symptoms of vascular stress were submitted to land-grant university plant diagnostic labs for disease diagnosis. Plant symptoms included general stunting, leaf chlorosis, wilting, marginal leaf scorch, limb dieback, stunted foliage at the base of dead limbs, and internal vascular discoloration in the form of thin streaks extending longitudinally within sap wood (Beckerman et al. 2022, Bily et al. 2023, personal communication Mike Munster). A fungus was consistently isolated from vascular streaks that had *Rhizoctonia*-like characteristics: no spore development in culture, large hyphae (5-8 µm in width), right-angle branching habit, and a constriction at the right-angle branch. The ITS region was sequenced and analyzed using the NCBI BLAST tool, which showed a 98.59% match with Ceratobasidium theobromae (Ct), a fungus associated with vascular streak dieback (VSD) in cacao (Theobroma cacao), a disease with symptoms that match our observations in maple and redbud (Guest and Keane 2007, Samuels et al. 2012). Further work is being conducted to determine if the Ceratobasidium species observed is a new species. Since 2019, Ceratobasidium has been detected in nursery stock or recent transplants in 12 states (AL, FL, IN, MD, MO, NC, OK, OR, PA, SC, TN, VA) in over 25 plant genera (Liyanapathiranage et al. 2024, personal communication Fulya Baysal-Gurel). External symptoms appear consistent across plant species, with wilting, marginal leaf scorch, and limb dieback. Internal symptoms may differ between plant species and within the same genus, with some species developing very distinct vascular streaking (C. canadensis) while others may have only a general discoloration within the wood (Cornus florida). In most cases, there are other disease and cultural problems contributing to general plant decline, which may initially confound diagnosis/detection of Ceratobasidium in symptomatic plants. These include problems such as improper planting depth, excessive irrigation, excessive fertilization, fungal root rot (Fusarium spp., Phytopythium sp. Phytophthora spp.), and fungal cankers (Botryosphaeria spp., Phomopsis spp., Cytospora spp.) (Fig. 1).

In VSD-positive plants, *Ceratobasidium* is observed almost exclusively within the xylem where vascular streaks are present. The vascular streaks are initially individual xylem vessel elements that contain hyphae consistent with *Ceratobasdium* and become occluded with a gumlike substance, consistent with a vascular infection (Figs. 2&3). Incubation of symptomatic tissue in a moist chamber to encourage fungal growth is one method used for detection. *Ceratobasidium* will grow out of the exposed xylem in large tufts of hyaline to yellow mycelium in as little as two days or as long as 20 days. *Ceratobasidium* can be isolated from symptomatic tissue onto artificial media; however, it is very slow growing and can take weeks to cover a plate. It is also difficult to keep the fungus alive on artificial media as successive culture transfers from

the original plate lead to reduced fungus vigor, and it eventually dies out (Liyanapathiranage et al. 2024). The speed and amount of growth is highly dependent on multiple factors, including the time of year the sample was collected, how the sample was maintained prior to incubation/plating (storage temperature), and whether the sample tissue is still moist or has dried out. Moisture appears to be a major factor in the overall growth of the fungus, and if samples are dry or old, it can take longer for *Ceratobasidium* to grow out of affected tissue, if at all. Similarly, there has been no success in isolating *Ceratobasidium* from some plant species, including *Cornus*, even when detected by PCR, which suggests that there is some variability in our ability to isolate the fungus from VSD-positive plants. Due to this complication, molecular testing methods are a great asset in being able to detect *Ceratobasidium* in VSD-symptomatic plants in plant samples that are of poor quality and are from species from which we cannot isolate the fungus. The inability to maintain cultures long term presents a significant roadblock for research efforts because it requires isolation of the fungus from living plant tissue for each new study that is conducted.



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- Beckerman, J. 2022. Vascular Streak Dieback of Redbud: What Plant Pathologists Know so far. Purdue Landscape Report 22-11. Retrieved December 13, 2024. <u>https://purduelandscapereport.org/article/vascular-streak-dieback-of-redbud-what-plant-pathologists-know-so-far/</u>
- Bily, D., Rodriguez-Salamanca, L., and Bush, E. 2023. Vascular Streak Dieback: An emerging problem on woody ornamentals in the U.S. Virginia Cooperative Extension. Retrieved December 13, 2024. <u>https://www.pubs.ext.vt.edu/SPES/spes-483.html</u>
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#### Vascular Streak Dieback of Woody Ornamentals in Virginia

Devin Bily<sup>1</sup>, Tashi Gyatso<sup>1</sup>, Farhat Avin<sup>2</sup>, Prabha Liyanapathiranage<sup>2</sup>, John Bonkowski, Lina Rodriguez Salamanca<sup>4</sup>, Fulya Baysal-Gurel<sup>2</sup>

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Vascular streak dieback (VSD) is a critical issue affecting woody ornamentals and the nursery industry in the U.S. A fastidious *Ceratobasidium* sp. (*Csp*) fungus has been associated with VSD, which has been confirmed in at least 11 states from over 25 broad-leaved woody host plant genera (Liyanapathiranage et al. 2024). This emerging problem has significantly affected the deciduous flowering and shade tree market in the U.S., valued at over \$1.8 billion in 2019 (National Agricultural Statistics Service 2020). *Csp* was first detected in Virginia in April 2022 from flowering dogwood nursery stock with symptoms of wilt and dieback, which coincided with reports from other states (Beckerman et al. 2022; Bily et al. 2023).

From April to November of 2022 to 2024, 997 plants were tested using PCR/qPCR from 86 host genera from 91 nurseries and 13 landscape sites across 53 municipalities in Virginia. The objective was to understand the distribution, host range, and impact of *Csp* in the ornamental nursery industry and determine the phylogenetic position of the Virginia *Csp* strains with strains from Tennessee and other closely related taxa.

In total, the fungus was detected in 285 of 997 symptomatic plants tested (28.6%), representing 45 broadleaf and two coniferous woody host genera from 53 nurseries and eight landscape sites across 37 municipalities in Virginia. *Csp* was confirmed in 174 plants using conventional PCR (61.1%) and 111 with qPCR (38.9%), with 65 ITS, 13 LSU, 5 TEF1- $\alpha$ , 5 ATP6, and 5 RPB2 sequences generated from the amplicons. *Csp* was detected most (53.5%) in *Cercis* (27% *n* = 77), followed by *Cornus* (15.4% *n* = 44), and *Acer* (11.2% *n* = 32). However, these genera accounted for 39.92% of the total samples tested. These results demonstrate that *Csp* is associated with many symptomatic woody plant hosts in Virginia nurseries, and that the strains from Virginia and Tennessee are genetically identical based on a five gene phylogeny.

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- Beckerman, J. 2022. Vascular Streak Dieback of Redbud: What Plant Pathologists Know so far. Purdue Landscape Report 22-11. Retrieved September 2, 2024.
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#### Vascular Streak Dieback Multi-State Survey Results Prabha Liyanapathiranage

#### Tennessee Department of Agriculture, Ellington Agricultural Center, Nashville, TN 37220

The Tennessee Department of Agriculture, in collaboration with various government agencies and academic partners, is conducting a comprehensive survey to assess the distribution and host range of Vascular Streak Dieback (VSD), a disease presumed to be caused by Ceratobasidium sp. This study is funded by the U.S. Department of Agriculture (USDA) Animal and Plant Health Inspection Service (APHIS) under the PPA7721 Plant Pest and Disease Management and Disaster Prevention Program. Survey participants were provided with a digital questionnaire and diagnostic materials to assist in VSD identification and sample handling, including shipping protocols. Data collection was facilitated using the Survey123 platform. Throughout the 2023-2024 survey period, samples were collected from 72 counties across eight states: Tennessee, Oregon, Maryland, Missouri, South Carolina, Alabama, Indiana, and Kentucky. In total, 406 samples were collected from 21 plant genera. Of these, 67% were from Cercis spp., 17% from Acer spp., and 8% from Cornus spp. The majority of samples (85%) were obtained from nurseries, while the remaining samples came from garden centers, retail and wholesale sellers, landscape suppliers, and landscapes. Samples were tested for the presence of Ceratobasidium sp., the suspected pathogen responsible for VSD. DNA extraction from plant samples was followed by the confirmation of *Ceratobasidium* presence using *Ceratobasidium*specific qPCR primers. Of the 406 samples, 96 (24%) tested positive for *Ceratobasidium* sp. Approximately 70% of the positive samples were from Cercis canadensis, with the remainder identified in a range of plant species, including Acer spp., Cornus spp., Magnolia spp., Ginkgo biloba, Liriodendron tulipifera, and Nyssa sylvatica. Notably, the recent identification of Pyrus communis (pear) and Rosa spp. (rose) as positive for Ceratobasidium sp. represents a significant expansion in the host range of this emerging pathogen. Symptomatic responses reported in the survey included dieback, vascular discoloration, leaf scorch, mouse-eared leaves, wilting, and the formation of epicormic shoots. This survey provides crucial data on the spread and impact of VSD, offering essential insights for the management of this novel and potentially devastating issue that threatens woody ornamental production.

#### National Needs Assessment Survey Results to Identify the Most Critical Research and Extension Priorities of Vascular Streak Dieback (VSD)

Prabha Liyanapathiranage<sup>1</sup>, Kumuditha Hikkaduwa Epa Liyanage<sup>2</sup>, Fulya Baysal-Gurel<sup>2</sup>

<sup>1</sup>Tennessee Department of Agriculture, Ellington Agricultural Center, Nashville, TN 37220 <sup>2</sup>Tennessee State University, College of Agriculture, Otis L. Floyd Nursery Research Center, McMinnville, TN 37110

The nursery industry is facing a serious threat from Vascular Streak Dieback (VSD), which has been identified in more than 25 ornamental and woody plant genera in multiple U.S. states since it was first identified in 2020. Eastern redbud has the most cases, followed by maple and dogwood. The primary goal of this survey was to identify stakeholders' most critical research and extension priorities in advance of the VSD Meeting sponsored with a USDA-NIFA-SCRI planning grant. A survey was conducted using a questionnaire to collect data via email from a list of potential meeting attendees. The majority of respondents were researchers (64%), followed by regulatory agency staff (12%), allied industry members (8%), and nursery owners (8%). The respondents averaged more than 16 years of nursery industry-related experience. All the respondents were well-aware of VSD and its symptoms, and 96% had observed VSD symptoms in nursery, landscape, forestry, and related environments. Redbud was identified as the major nursery crop exhibiting VSD symptoms over the last five years, followed by dogwood and maple. Respondents identified VSD as the issue having the most impact on the saleable quantity and quality of nursery crops, followed by phytophthora, canker, and pythium diseases. Respondents identified buds/liners/cuttings, cutting and pruning tools, plant debris, nursery equipment, and other crops as the most important nursery production practices in limiting/preventing VSD. They named university extension specialists as their preferred source of assistance on VSD-related problems, followed by county or regional extension officials, government agencies, and growers. Asked what is most important to them, respondents named information about methods to prevent and manage VSD as their top preference, followed by disease-management products, understanding disease sources, and disease identification. Respondents' preferred methods for receiving information were websites, webinars, factsheets, grower meetings, and field days (vs. emails, social media, website-based community groups, and magazines). This survey was important in identifying research areas discussed at the VSD Meeting and in developing objectives for the USDA-SCRI pre-proposal submission "Managing Vascular Streak Dieback: A Collaborative Effort to Understand and Mitigate the Rising Threat in Woody Ornamental Crops."

#### Economic Impact of Vascular Streak Dieback (VSD) Threat of Redbud in the U.S. Kumuditha Hikkaduwa Epa Liyanage and Fulya Baysal-Gurel

Tennessee State University, College of Agriculture, Otis L. Floyd Nursery Research Center, McMinnville, TN 37110

The USDA 2022 Census of Agriculture reports that nursery crop sales totaled about \$8 billion, spanning around 19,000 farms and covering over 401,000 acres (USDA-NASS, 2022). Additionally, the USDA's 2019 Census of Horticultural Specialties identifies redbuds as the fifth most valuable deciduous flowering tree crop in the U.S. (USDA-NASS, 2020). Recently, the U.S. redbud production industry has faced a major challenge from a new threat known as Vascular Streak Dieback (VSD). The disease causes significant losses for growers by deteriorating plant quality and marketability, as well as raising labor costs linked to essential management practices (Liyanapathiranage et. al., 2024). The primary goal of this project is to thoroughly understand redbud production practices and conduct an in-depth analysis of the economic impacts of VSD to effectively address the specific needs of the U.S. redbud nursery industry. A multi-state survey was conducted to collect data from stakeholders involved in the redbud industry in 2023. Forty-eight responses were received from seven states including Tennessee, Virginia, North Carolina, Oregon, Ohio, Oklahoma, and Kentucky. In redbud operations, container production was the focus for the majority of growers, followed by ball and burlapped, bareroot, and liner production. In recent growing seasons, 77% of sampled growers reported VSD symptoms in redbud plants. VSD issues have been noted over the last five years, with 2022 having the highest incidence rate at 30%. Symptoms were observed most frequently in summer (37%), followed by spring, fall, and winter. VSD accounted for most redbud loss, affecting around 70% of production, followed by winter damage, root rot, and other reasons. On average, growers reported VSD losses of around 25% in container plants, as compared to other redbud production stages. Key factors in predicting future redbud production behavior included having over 30 years of nursery experience, nursery size, redbud landscape plants exhibiting VSD symptoms, the extent of redbud container production, and VSD-related economic losses.

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#### Detection of *Ceratobasidium* sp. (*Csp*) Associated with Vascular Streak Dieback (VSD) of Woody Ornamentals in the United States Farhat Avin<sup>1</sup> and John Bonkowski<sup>2</sup>

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*Ceratobasidium* sp. (*Csp*) associated with vascular streak dieback (VSD) represents a significant diagnostic challenge due to its complex symptomatology and genetic diversity. This paper expands on the molecular diagnostic challenges and introduces a suite of novel genomic and molecular tools developed to enhance the detection and understanding of this pathogen.

Several new molecular markers were developed, and advanced genomic techniques were employed to improve the specificity and sensitivity of *Csp* detection. The main focus was the introduction of the CP\_qP\_Csp primer set for PCR and qPCR assays, complemented by a discussion on the selection and optimization of additional genetic markers including ITS, LSU, RPB2, TEF1, and ATP6 for broader genomic studies.

Through ongoing whole-genome sequencing and phylogenetic analysis using concatenated sequences from multiple genetic regions, we began exploring the complex genomic structure of *Csp*. This included its evolutionary relationships with closely related species and its genetic adaptations to different hosts and environments. These studies have provided critical insights into the pathogen's virulence factors and its interactions with host plants.

In addition to the CP\_qP\_Csp primer pair, other markers were developed and optimized to improve diagnostic accuracy. These efforts included refining existing primers and designing new ones that target less-conserved genomic regions, enhancing the ability to detect and differentiate *Csp* even in mixed microbial communities.

The new CP\_qP\_Csp primer pair demonstrated exceptional performance, with sensitivity levels reaching as low as 300 fg/ $\mu$ L in qPCR assays. The broader marker set also showed high specificity in distinguishing *Csp* from closely related pathogens. Cross-platform validation confirmed the effectiveness of these markers across different diagnostic settings by significantly showing low false positive and negative results.

Extensive validation was conducted in collaboration with several academic and governmental diagnostic institutions such as Purdue University, the Virginia Department of Agriculture and Consumer Services, Virginia Tech University, Oklahoma State University, Ball Horticultural Company, and Bartlett Tree Experts. This collaborative effort not only confirmed the robustness of the assays but also facilitated the integration of these new tools into routine diagnostic workflows, ensuring broad applicability and practicality.

The development of these molecular and genomic tools significantly advances the capabilities in diagnosing and further managing VSD. By integrating these advanced techniques into current practices, more accurate and timely diagnoses can be offered, which are crucial for effective disease management and mitigation strategies.

#### Koch's Postulates and/or Hill's Criteria: Progress on Assessing the Relationship Between *Csp* and VSD Prabha Liyanapathiranage<sup>1</sup> and Fulya Baysal-Gurel<sup>2</sup>

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Determining the pathogenicity of *Ceratobasidium* sp. (*Csp*) is essential for confirming its role in vascular streaking and dieback associated with Vascular Streak Dieback (VSD). It remains unclear whether Csp is the primary causal agent of VSD or an opportunistic pathogen that becomes pathogenic due to plant stress caused by other pathogens or environmental factors. Koch's postulates have not been fully satisfied due to challenges in isolating and maintaining Csp cultures. To address this, healthy Eastern redbud plants were inoculated with Csp through leaf and stem inoculation techniques. Vascular streaking symptoms appeared two months after stem inoculation; however, re-isolation of Csp from infected tissue was unsuccessful, preventing the completion of Koch's postulates. Similarly, Koch's postulates have not been fulfilled for Ceratobasidium theobromae (Ct) as the causal agent of VSD in cacao, although strong epidemiological evidence supports its association with the disease. Pathogenicity tests on cacao have demonstrated that inoculation with Ct induces typical VSD symptoms, such as chlorosis and tissue discoloration. However, additional research is necessary to determine whether VSD in multiple plant species is caused solely by Ct or if co-infection with other pathogens plays a role. Pathogenicity tests on other fungi, including Neopestalotiopsis sp., Fusarium sp., Diaporthe sp., Didymella sp., and Botryosphaeria sp., showed that while these fungi caused vascular discoloration and lesions, they did not produce the characteristic marbled wood appearance associated with VSD. It is hypothesized that *Csp* may weaken plants, allowing other pathogens to invade and exacerbate VSD symptoms. Similar interactions have been observed in cacao, where Fusarium, Colletotrichum, and Lasiodiplodia spp. are linked to VSD. In conclusion, while the pathogenicity of Csp as the primary cause of VSD remains inconclusive, evidence suggests it may contribute to the development of VSD-related symptoms under specific conditions. The difficulty in re-isolating Csp from infected tissue indicates it may act as an opportunistic pathogen rather than the sole causal agent. Co-infection with other pathogens, such as Fusarium and Botryosphaeria, may provide further insights into VSD development. Continued research is essential to better understand the interactions between Csp and other fungal pathogens and to identify the specific conditions under which Csp becomes pathogenic. This knowledge will be crucial for developing effective management strategies for VSD in woody ornamentals.

#### Predicted Biology and Epidemiology: Environmental Factors and Seasonality of VSD Symptom Development and Spread; Preliminary Results from TSU's Spore Trapping Trials; Potential for VSD Forecasting System Prabha Liyanapathiranage<sup>1</sup> and Fulya Baysal-Gurel<sup>2</sup>

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Vascular streak dieback (VSD) is an emerging concern in the production of woody ornamentals in the U.S., with little available information regarding its biology and epidemiology within this specific context. To gain a deeper understanding of this issue, it is crucial to review the existing literature on VSD in other host plants and explore potential correlations with the current situation in the U.S. VSD was first identified in cacao in New Guinea in the 1960s and has since spread to various cacao-growing regions across Asia, including Indonesia, Malaysia, and the Philippines. The disease is caused by Ceratobasidium theobromae (Ct), a pathogen believed to have evolved as an asymptomatic endophyte in native Southeast Asian plants before adapting to cacao. The disease in cacao begins with chlorotic leaves that develop necrotic blotches and green islets within 3-5 months after infection. As the disease progresses, chlorosis spreads to adjacent leaves, leading to defoliation. Infected leaves tend to remain attached longer than healthy ones, facilitating sporulation of Ct on the leaf veins and petioles. Ct basidiospores germinate on young cacao leaves, colonizing the xylem tissue, which causes vascular browning and necrosis. This eventually leads to dieback, as the pathogen spreads from the leaves to branches and stems, sometimes killing entire branches or the whole tree, particularly in younger cacao plants. Infected leaves, once they fall, release white corticioid basidiocarps, which produce basidiospores that are wind-dispersed over short distances. The progression of the disease and the severity of symptoms are closely linked to environmental factors, particularly rainfall. Prolonged wet periods are essential for the formation and discharge of basidiospores, creating conditions that promote disease spread by increasing the availability of free water, which is necessary for both spore germination and the infection of healthy plant tissue. Preliminary research conducted by Tennessee State University (TSU) in 2023 aimed to understand VSD symptom development in relation to weather conditions. Weather stations were installed at commercial nurseries producing redbud trees in Tennessee, recording parameters such as minimum and maximum temperature, solar radiation, rainfall, and relative humidity from June to November. Redbud plants were assessed for VSD symptoms during this period. The results indicated that prolonged rainfall and low solar radiation likely contributed to the accelerated symptom development observed during the later months. These findings align with similar patterns observed in cacao, where extended rainy periods have been associated with the development of VSD symptoms in young plants. In Eastern redbud seedlings (1 year old), VSD symptoms typically begin to appear by June, as new leaves emerge. As the disease progresses, the leaves gradually become chlorotic, with leaf scorching becoming more evident from July to September. The scorching begins in individual leaves and spreads to entire branches. By October and November, heavy infection can lead to the death of the entire plant. While there is no evidence to support seed transmission, key questions persist, such as the viability of spores, the potential vascular movement of mycelium from graft unions, the presence of a dormancy period, and how various management practices impact disease spread and symptom development. Spore trapping is a critical technique for identifying and quantifying pathogenic spores in field

conditions, providing valuable insights into the disease cycle. TSU has used passive deposition traps to detect fungal spores in areas where VSD is naturally present. These early findings identified *Ceratobasidium* spores in redbud fields, and the detection coincided with management practices such as pruning and seasonal defoliation at the end of the growing season. These results highlight the importance of ongoing research into spore detection, which could be pivotal in developing a VSD forecasting system and more effective management strategies.

#### **Current and Novel Management Strategies and Challenges**

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Vascular streak dieback (VSD), first identified five years ago in eastern redbud in Tennessee and red maple in North Carolina, has become a major concern for nurseries across multiple states, including Tennessee, Virginia, and North Carolina, with isolated cases in Indiana, Florida, and Oklahoma. VSD has been detected in newly planted landscapes, botanic gardens, and natural areas, significantly impacting eastern redbud and more than 25 other woody ornamental plant genera. The economic impact on redbud, red maple, and dogwood alone is estimated to exceed \$175 million.

VSD symptoms vary by host and include stunted growth, yellowing, dieback, leaf scorch, vascular streaking, and poor root development, which can mimic other conditions like Verticillium wilt, bacterial leaf scorch, and laurel wilt. Co-occurring pathogens, such as Botryosphaeria canker fungi, often complicate diagnosis. The disease has caused significant losses for the nursery industry, with rejected shipments, canceled orders, and widespread destruction of symptomatic and asymptomatic plants due to Stop Sales orders.

Currently, no definitive chemical treatments exist for VSD. However, recognizing the urgency of assisting nursery producers, Tennessee State University (TSU) collaborated with industry partners to conduct fungicide efficacy trials in 2022 using redbud plants naturally exhibiting VSD-related symptoms. The results indicated that foliar applications of Postiva (FRAC 3 + 7) at 20 fl oz/100 gal and Mural (FRAC 7 + 11) at 7 oz/100 gal at 14-day application intervals were the most effective treatments in reducing leaf scorch associated with VSD on eastern redbud seedlings, on three-year-old plants, and on several two-year-old budded cultivars growing in field and container settings. These treatments also reduced the population levels of canker-causing pathogens such as *Botryosphaeria* and *Didymella*. Although some treatments were identified as effective in reducing symptoms, they did not cure already-infected plants. TSU is continuing its work to identify more effective chemical treatments.

Another research focus is identifying VSD-tolerant redbud cultivars. Screening trials revealed that cultivars with yellow foliage and papery leaves are more susceptible, while those with dark green or purple foliage and leathery leaves exhibit greater tolerance. This information is critical for breeding resistant hybrids. The TSU team and Dr. Hsuan Chen's team at North Carolina State University are continuing with the cultivar screening, including more redbud species, cultivars, and hybrids, to identify VSD resistance, funded by the North Carolina Department of Agriculture and Consumer Services and the Horticultural Research Institute.

#### Ornamental Plant Breeding: Cercis Breeding and VSD Hsuan Chen

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The VSD pathogen, *Ceratobasidium* sp. (*Csp*), has been identified in over 23 ornamental plant species (Baysal-Gurel, 2023), with eastern redbud (Cercis canadensis), dogwood, and maple tree productions being the most severely affected. Identifying genetic resources and breeding for VSD-resistant cultivars represents the ultimate long-term solution. Fortunately, redbud species-specific VSD tolerance/resistance has been primarily observed, showing great potential in eastern redbud breeding. This observation was primarily confirmed in a 1.5-year trial at Tennessee State University. North Carolina State University (NCSU) has a long history and repetition in Cercis breeding, and North Carolina has been one of the top three states to produce Cercis. Since 2022, several nurseries in the state have reported to NCSU that an Asian species, C. chinensis, has displayed strong tolerance/resistance. Several years ago, NCSU developed two adult phase F1 interspecific hybrid plants (C. chinensis x C. canadensis). These F1 hybrid plants are now in the adult phase, and their proven fertility has successfully produced BC1 plants. We are working on introducing the VSD resistance gene from C. chinensis into C. canadensis; in the meantime, we are working to discover additional potential VSD resistance from other Cercis species, including C. occidentalis, C. siliquastrum, C. racemosa, C. glabra, and different C. chinensis accessions. Our target is breeding and recommending VSD-resistant cultivars or plant materials for the nursery industry.

#### Understanding the Role of Container Color and Root Rot Pathogen (*Phytopythium vexans* L.) in Predisposing Redbuds to Vascular Streak Dieback Madhav Parajuli<sup>1</sup>, Jake Shreckhise<sup>1</sup>, Cansu Oksel<sup>2</sup>, and Fulya Baysal-Gurel<sup>2</sup>

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Redbud (*Cercis canadensis* L.), known for its attractive flowers and heart-shaped leaves, is an economically important and popular ornamental landscape tree widely cultivated across the U.S. (National Agriculture Statistics Service 2020). Tennessee ranks first nationally in redbud sales, with approximately 228,661 trees sold annually, representing 31% of all redbuds sold in the U.S. (National Agriculture Statistics Service 2020). In recent years, the production and sale of redbuds have been greatly impacted by vascular streak dieback (VSD), a disease caused by a newly encountered fungal pathogen *Ceratobasidium* sp. D.P. Rogers (Csp) (Liyanapathiranage et al. 2024). Leaf scorch, branch dieback, and vascular streaking are the most common symptoms of VSD. While much remains unknown about VSD and its causal agent, various biotic and abiotic factors are likely associated with its severity and progress and must be further explored.

Conventional black plastic nursery containers absorb solar radiation, resulting in daytime root-zone temperatures (RZTs) that can exceed 50°C during the summer months (Ingram et al. 2015). Root growth of many woody landscape plants ceases as RZTs rise above 38°C, and exposure to >46°C can kill root cells within 20-30 mins (Ingram et al. 2015). When RZTs are supraoptimal but do not directly injure roots, physiological processes such as photosynthesis, water uptake, nutrient absorption, and hormone signaling may be severely impaired. This systemic stress caused by supraoptimal RZT can, in turn, predispose plants to root rot diseases (Krebs, 2016). Using white plastic nursery containers, which reflect instead of absorb solar radiation, has repeatedly been shown to reduce RZT by up to 10°C, resulting in faster-growing plants with superior root systems (Witcher et al. 2020). Additionally, root rot pathogens are known to weaken trees' defense systems. The combined physiological stress of elevated root-zone temperatures in black containers and infection by root rot pathogens is hypothesized to increase the susceptibility of redbuds to VSD.

A field study was conducted at the Otis Floyd Nursery Research Center in McMinnville, TN to examine the impact of container color (i.e., white vs. black) and the root rot pathogen *Phytopythium vexans* on predisposing eastern redbuds to VSD. The trial was conducted next to a plot of redbuds with confirmed VSD. On May 20, 2024, 32 one-year-old eastern redbuds were potted in 2-gallon (PF800) white containers, and an equal number of redbuds were potted in 2gallon (PF800) black containers. After three days, half of the plants in each container color were drenched with the biocontrol agent, RootShield® Plus WP (*Trichoderma harzianum Rifai* + *T. virens;* 0.6 g/L), and the application was repeated every six weeks. Ten days after potting, *P. vexans* was drench inoculated in half of the plants, both with and without the biocontrol treatment. Treatments were arranged in a completely randomized design with eight replications. Plants were irrigated using spray stakes. Irrigation application volume was adjusted biweekly to maintain a 20-30% leaching fraction based on replacement of daily water use/loss from the previous 24 h period. The trial ran for four months, from May 20 to September 20, 2024. Plant growth index (height and width) and leaf chlorophyll content were measured every three weeks, beginning one week after planting. Plant caliper was measured six inches above the

substrate level. Plants were also evaluated for leaf scorch, branch dieback, and vascular streaking. At the end of the trial, roots were evaluated for root rot severity (% roots affected). Plants grown in white containers had a significantly greater caliper, width, and chlorophyll content compared to those in black containers, while plant height remained unaffected by container color. The maximum daily RZT was, on average, 4°C lower in white compared to black containers. As such, the larger plant size observed in white containers is attributed to lower RZTs, which is consistent with findings from other studies that showed improved plant growth in white containers (Markham et al. 2011; Witcher et al. 2020). Root-zone heat stress was particularly evident along the southern and western container walls within black containers, where roots were either dead or absent. Neither the root rot pathogen nor the biocontrol agent affected height, width, or caliper. The biocontrol agent significantly reduced both root rot severity and leaf scorch progression. We did not find differences in final leaf scorch or vascular streaking between pathogen-inoculated vs. non-inoculated plants. Moreover, container color did not affect root rot severity or leaf scorch. While this research should be repeated before making definitive conclusions, these preliminary results are in opposition to our hypothesis that redbud produced in white containers have reduced root rot disease and VSD severity.

Although there are limited studies on this topic, previous studies have reported increased root rot severity in plants grown at higher RZTs. For instance, rhododendron (*Rhododendron sp.*) and hibiscus (*Hibiscus rosa-sinensis*) exposed to RZTs >40°C exhibited greater root rot severity from infections caused by *Phytophthora cinnamomi* and *P. nicotianae*, respectively, compared to those grown at lower RZTs (Krebs, 2016; Lyles et al., 1992). Redbud, compared to other temperate tree species, is tolerant of high RZTs (Markham et al. 2011), which may partially explain why root rot severity did not differ between plants grown in white vs. black containers. Also, the study site likely had limited VSD pathogen pressure due to its lack of a history with redbud production. The study will be repeated in summer 2025.

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#### Environmental Modification to Induce or Reduce Disease: Irrigation and Air Circulation as a Vehicle for IPM Practitioners and a Tool for Researchers Amy Fulcher

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A susceptible host, the infectious microorganism, and a conducive environment compose the disease triangle, the classic model for explaining the interactions of these factors on disease (Agrios 2005). Environmental modification, specifically moisture management, through both irrigation, i.e., scheduling and delivery practices, and air circulation can be an important part of an IPM program by altering the environmental leg of the disease triangle. Environmental modification, naturally or through intervention practices, can limit fungal-borne diseases, potentially including *Ceratobasidium* sp. (*Csp*), the putative cause of vascular streak dieback among ornamental crops in the U.S. Thus, cultural practices that modify the production environment should be considered as IPM approaches to preventing or limiting incidence and severity of *Csp* and its spread in nursery crops.

Conversely, environmental modification may also be used to induce infection. Humidity chambers are often utilized when establishing or maintaining fungal colonies in laboratories. In outdoor settings, environmental modifications may be a tool to induce infection in experiments and to more rapidly and more assuredly determine the effect of treatments. For example, 1) irrigation practices that increase the frequency and longevity of leaf wetness and 2) production practices and site characteristics including plant spacing and natural and artificial windbreaks that impede air flow can be used to create the conditions that promote sporulation, spore germination, and infection in outdoor environments, and to influence disease spread. Scientists can manipulate these environmental conditions to induce infection when testing fungicides, cultural practices, or resistant varieties, for example, by "tipping" the environmental aspect of the disease triangle in favor of the pathogen.

Irrigation scheduling is traditionally manipulated to prevent water deficit, plant stress, and simultaneously maximize (or at least not compromise) crop health and growth. But irrigation can also be managed to induce water deficit and thereby control growth in lieu of plant growth regulators (Bayer 2020a, 2000b, 2000c). Reduced irrigation can also be used to induce water deficit in order to test the effect of water stress on disease development (Beaulieu et al. 2022; Del Castillo Múnera et al. 2019; Neupane et al. 2022).

Irrigation scheduling logic that can be used to manipulate plant water status may be plantbased or substrate-based. Automated systems coupled with sensors make on-demand scheduling logic possible (Fulcher et al. 2012). Some examples of logic on which to base nursery irrigation scheduling include maximizing photosynthetic rate (Fulcher et al., 2012; Hagen et al. 2014; Basiri Jahromi et al. 2018), maximizing plant available water (Basiri Jahromi et al. 2020a), daily water use (Pershey et al. 2015), leaching fraction (Cypher et al. 2022) and evapotranspiration (Million and Yeager 2015). These logic-based systems are recognized for their ability to maintain healthy crops and acceptable growth rates and to conserve water.

In contrast, static irrigation systems, such as those that are timer-based or manually operated at the same time and for a set duration, do not respond to environmental change or crop changes, i.e., growth, pruning, that elicit a greater or lesser evapotranspirational demand, and thus are prone to over- or under-irrigating. Manual irrigation is also subject to employee error due to customer interruptions, responsibilities related to other tasks, and other forms of human per irrigation event but were irrigating 0.3-1.3 inches per day – a 4.3-fold difference in amount of irrigation (Fare et al. 1992). In an informal survey of Tennessee nurseries manually operating irrigation, leaching fraction, the portion of irrigation water applied that leaches from the container, was as high as 94% (Fulcher, unpublished data). Static irrigation, unless closely and carefully managed, can lead to extremes of too wet or too dry. The following are two case studies that illustrate the potential to use environmental modification as part of an IPM program to reduce fungal-borne disease. **1. Using Fans to Prevent Downy Mildew in Basil** Sweet basil is plagued by downy mildew (Cohen and Ben-Naim 2016). Basil crops, especially in warm, humid conditions, are particularly affected by downy mildew. The causal agent, *Peronospora belbahrii*, an oomycete, sporulates at night with 7.5 or more hours at a

relative humidity of 95% or greater and can infect leaves with as little as four hours of moisture on the leaf surface (Cohen and Ben-Naim 2016). Nocturnal fan use can prevent downy mildew in basil by circulating air and reducing moisture around plants (Cohen and Ben-Naim 2016). Cohen and Ben-Naim (2016) found that nocturnal fan use decreased the amount of time with 95% or greater relative humidity, which limited dew formation and reduced both sporulation and infection. Cohen et al. (2017) found that

error. Poor irrigation system infrastructure can exacerbate the effects of human error. For

example, Alabama nurseries using timers or manual irrigation intended to apply 1 inch of water

using a relative humidity sensor – to activate the fans when the relative humidity reached 70% and to deactivate them when it dropped back down to 65% – was effective and saved electricity. Omer et al. (2021) reduced downy mildew by up to 73% by operating fans once every hour for 15 minutes from 7 p.m. until 8 a.m.

#### 2. Using Fans to Prevent Fungal-Borne Disease in Golf Course Greens

Golf course greens are maintained in a similar fashion to nursery crops: 1) they are grown in modified soil or soilless substrate, 2) abundant amounts of water and fertilizer are applied, and 3) growth is controlled mechanically via sometimes aggressive and/or frequent pruning or mowing that creates wounds. In these systems, when overhead irrigation is used or dew forms, very tender growth, damaged leaf tissue, or both, experience long periods of leaf wetness – conditions that foster fungal sporulation and infection. Many U.S. golf courses in the South, the transition zone, and even the Northeast (Moeller and Chapin 2011) use air circulation as part of an IPM program to prevent fungal-borne disease.

Fans can potentially lower disease by reducing the duration of leaf wetness, thereby preventing sporulation and influencing spore dispersal – preventing a concentration of spores. Peacock and Lyford (2022) tested fans operated at 2.7, 1.6, 0.7, and 0.3 m s<sup>-1</sup> above ambient wind speeds over greens that were surrounded by dense vegetation in North Carolina. They found that as air speed increased, duration of leaf wetness decreased. Additionally, disease was lowest at the highest air speeds (<7.3%) and greater at the low air speeds ( $\geq$ 26%). Similarly, Taylor (1995) found that air speed should be maintained between 1.4 to 2.6 m s<sup>-1</sup>. Lower air speed is ineffective at preventing disease, and a greater wind speed can desiccate the crop plant.

Using fans to increase air circulation can also modify the temperature. Increasing air circulation cools the canopy during hot, humid periods by increasing evapotranspiration, and potentially reduces crop stress. Peacock and Lyford (2022) found that the turf canopy temperature was 3.5 to  $5.5^{\circ}$ C hotter with  $<0.3 \text{ m s}^{-1}$  wind speed compared with 2.7 m s<sup>-1</sup>.

#### Extension and Outreach Efforts in Tennessee and Beyond For Vascular Streak Dieback Research Kaitlin Barrios

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Nursery stock sales in Tennessee totaled approximately \$201 million and ranked 10<sup>th</sup> in the nation in 2022 according to the United States Department of Agriculture (USDA) Census of Agriculture. Tennessee nursery stock is shipped throughout the U.S. and has the potential to bring with it many fungal or bacterial plant pathogens. In recent years in Tennessee, wilting and dieback have been observed in many redbud (*Cercis canadensis* L.) trees along with other symptoms. Symptoms of Vascular Streak Dieback (VSD) in redbud have been reported in neighboring states since the initial sample was submitted in 2019 to the Tennessee Department of Agriculture.

Much research has been conducted on VSD by pathologists and their teams at Tennessee State University, Purdue University, the Virginia Department of Agriculture and Consumer Sciences, and other agricultural research organizations to understand the causal agent(s) and investigate management options for growers, landscapers, and homeowners. There is still more to be understood about VSD and much remains to be done: identifying its causal agent(s) and how it is spread; trialing species and cultivars for resistance; identifying more host plants; and evaluating best-management practices. In October 2024, researchers, growers, diagnosticians, and business leaders convened in McMinnville, TN to hear updates, gain insights, and discuss plans for furthering the research and management strategies of VSD. To that end, the VSD Working Group has composed a pre-proposal to apply for a USDA National Institute of Food and Agriculture (NIFA) grant award to support these continued efforts. Thus far, research findings have been shared via in-person and virtual presentations, scholarly publications, extension fact sheets, industry magazines, scientific posters, and more. The VSD Working Group plans to disseminate findings and updates through the aforementioned channels, as well as social media platforms, grower workshops and/or trainings, and a website hosted by the Horticultural Research Institute (HRI). The website would serve as an informational hub, providing access to pictorial guides of VSD symptoms, a list of diagnostic labs for testing samples, an Early Detection and Distribution Mapping System (EDD MapS), and additional resources. The Outreach and Extension team also plans to provide resources in both Spanish and English. Current collaborators plan to provide training in their regions within North Carolina, Oregon, Tennessee, and Virginia; we hope to partner with extension and/or educational personnel in additional states in the future. Due to the current and anticipated impacts of VSD on the ornamental plant industry, many collaborators are investing their time and effort to tackle this problem as soon as is feasibly possible.

#### Sustainable Solutions for VSD: Leveraging Education, Outreach, and Collaboration Lana Petrie

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Dr. Lana Petrie will lead an initiative to develop and refine a comprehensive communication plan and curriculum designed to enhance outreach, education, and effective stakeholder engagement in agricultural development. The project focuses on fostering collaboration with partners, stakeholders, farmers, and students while leveraging communication tools developed with NIFA funding, including the Students Exploring Employment and Development (SEED) app, to deliver research-based, visually engaging content. The communication content and plan will be periodically updated and refined based on ongoing feedback to ensure continued relevance. Communication channels like the SEED app will remain integral to effectively connecting with stakeholders and students and disseminating educational materials. A sustainability strategy will be developed to ensure the communication plan's longevity and ongoing effectiveness. This initiative aims to provide impactful, up-to-date resources that advance education, outreach, and stakeholder relationships, fostering continued engagement and growth in agricultural development.

### **Identified Priorities and Critical Needs**

#### I. Identification & Diagnostic Testing

- 1) Improved culturing protocols
  - Long-term *Csp* storage/tissue culture
- 2) Genomic comparison
  - Compare the genome of *Csp* to closely related fungi
  - Understand disease mechanisms
  - Develop molecular assays
- 3) Pathogen spread and adaptation
  - Analyze genomes of pathogen samples from different hosts and locations
- 4) Inoculation experiments using *Csp* on redbud, flowering dogwood, and red maple
  - Assess effects of abiotic stress factors on symptom development
  - Develop ability to re-isolate *Csp*
- 5) Endophytic colonization
  - Explore whether *Csp* acts as an endophytic colonizer and transitions to an opportunistic pathogen
- 6) Molecular detection
  - Develop molecular tools to detect *Csp* in asymptomatic hosts
  - Understand colonization patterns
- 7) Disease management
  - Integrated disease-management practices using ecological & epidemiological insights

#### II. Biology/Epidemiology

- 1) Identify life cycle stages
  - Establish experiments to identify all unknown life cycle stages of VSD
- 2) Determine environmental parameters
  - Identify exact environmental parameters for infection, disease development, inoculum production, and plant responses by:
    - Measuring plant water potential
    - Determining VSD-specific volatile markers
    - Observing *Csp* colonization and localization within tissues
    - Using electron microscopy
- 3) Examine sporulation factors:
  - Study the effects of host phenology, time of day, temperature, and moisture on *Csp* sporulation of basidiospores
  - Understand conditions that lead to new VSD infections
- 4) Develop weather-driven VSD forecast model
  - Create a forecast model based on weather data
  - Host at USPest.org
  - Predict VSD outbreaks
- 5) Conduct spore-trapping studies
  - Drone method Best height for drone (5-10 m)
  - Stationary methods Burkard spore trapping 24hrs/7days
  - Weather stations
  - Field and cold storage

#### **III. Best-Management Practices**

- 1) Innovative monitoring and control tools
  - Develop new products and technologies
- 2) Resistant cultivars and production approaches
  - Identify and develop new cultivars that are more VSD resistant
  - Improve production approaches for pruning, fungicide application, fertilizer rates, and irrigation techniques
- 3) Breeding and marker-assisted selection
  - Focus on highly tolerant germplasm and cultivars from redbud, flowering dogwood, and maple collections
  - Use marker-assisted selection for breeding and gene verification
- 4) Redbud breeding and cultivar screening
  - Screen and confirm VSD tolerance in Chinese redbud (*C. chinensis*) and varying susceptibility in Eastern redbud (*C. canadensis*)
  - Develop F1 interspecific hybrid plants and BC1 plants
- 5) Rootstock comparisons
  - Compare susceptibility of Eastern redbud cultivars grafted on rootstocks of 2 species
  - Investigate the effectiveness of using *C. chinensis* as rootstock
- 6) Propagation methods
  - Compare growth rates and *Csp* presence in *in vitro* propagated and traditionally grafted liners
  - Evaluate tissue-cultured liners versus traditional grafting
- 7) Host resistance in maple and dogwood
  - Characterize host resistance in maple and dogwood
- 8) Disease mitigation
  - Identify critical control points at production nurseries
  - Enhance BMPs (Best Management Practices).
  - Evaluate new fungicides, biofungicides, and nanotechnology-based adjuvants
- 9) Improved nursery practices
  - Investigate improved practices
  - Container and field-grown nursery production
  - Evaluate irrigation, fertilizer application, leaf wetness prevention, sun exposure, and pruning
- 10) UAS-based remote sensing
  - Use Unmanned Aircraft System (UAS) for remote sensing
  - Monitoring crop conditions
  - Collect and analyze data using RGB
  - Multispectral cameras, LiDAR
- 11) Identification of effective VSD treatments
  - Assess different U.S. regions, multisite results

#### **IV. Economics**

- 1) Assess economic losses
  - Survey landscape companies, retail centers, and consumers to evaluate postproduction economic losses due to VSD
- 2) Economic analyses

- Perform economic analyses comparing BMPs to current management practices
- Calculate the net present value of long-term R&D in resistant/tolerant plants
- Consider other benefits of disease mitigation
- Cost effectiveness of new production practices
- Consumer preference and acceptability of VSD treatments
- 3) Consumer awareness and response
  - Conduct 2 studies to assess consumer awareness of VSD and their response to BMPs
  - Online survey
    - Assess consumers' interest in purchasing the crops impacted by VSD
    - Current purchasing behavior; VSD awareness; interest in/response to control methods
    - Perform conjoint analysis to capture how different product attributes influence consumer choice
    - Willingness-to-pay (WTP) for plants with those attributes
  - In-lab eye-tracking study
    - Key findings from the online survey (e.g., signs, labels, information nudges)
    - SmartEye Aurora stationary eye-tracking system
    - Visual attention metrics assessment

#### V. Outreach/Extension

- 1) Develop/maintain website with information produced by project team and affiliates
  - Post latest news and reports, pictorial guides to support VSD identification
  - Share videos, fact sheets, resources from collaborating institutions and organizations
  - Develop, market, manage, and host outreach webinars (quarterly) and videos online
  - Create and post project newsletters and other outreach materials
- 2) Use services such as EDDMapS/AgPest Monitor to report VSD, follow up with reporters, validate data, and produce maps based on available data
  - Assist with integrating maps or other data-based visualizations
  - Act as a sounding board for communication and evaluation plans of the project
  - Develop social media content, fact sheets, pamphlets, and other published materials and in-person training, in English and Spanish
- Collaborate with professional organizations (AmericanHort, ISPP, IPPS, the International Society of Horticultural Science, APS, NPDN, and the American Society for Horticultural Science) on programming for their conferences, seminars, and workshops
- 4) Create digital train-the-trainer program for extension agents and Extension Master Gardener volunteers
  - Provide guidance about topics such as identification, management, and plant selection
  - Make accessible on the Canvas learning platform
- 5) Develop short, engaging videos aimed directly at the public through consumer-focused channels and state platforms

# **Meeting Participants**

Last Name	First Name	Affiliation
Addesso	Karla	Tennessee State University
Adcock	Tracy	Scenic Hills Nursery
Alexander	Lisa	USDA-ARS
Averitt	Ben	Willoway Nurseries
Avin	Farhat	Tennessee State University
Barrios	Kaitlin	Tennessee State University
Batson	Alex	Ball Horticultural Company
Baysal-Gurel	Fulya	Tennessee State University
Bec	Sladana	Ball Horticultural Company
Bily	Devin	Virginia Department of Agriculture
Bienapfl	John	USDA-APHIS
Boggess	Sarah	University of Tennessee
Bonkowski	John	Purdue University
Borzouei	Shahla	Tennessee State University
Brown Jr.	Chris	Lancaster Farms
Calabro	Jill	Valent
Chang	Anjin	Tennessee State University
Chen	Hsuan	North Carolina State University
Chong	Juang-Horng (JC)	SePRO Corporation
Clendenon	Gary	Tennessee Department of Agriculture
Collier	Frank	Pleasant Cove Nursery
Creswell	Tom	Purdue University
Dawadi	Sujan	Tennessee State University
Driver	Jessica	Virginia Department of Agriculture
Erickson	Elizabeth	Knight Hollow Nursery
Fare	Donna	Horticulturist
Flanders	Paul	Botanico
Fulcher	Amy	University of Tennessee
Harmon	Carrie	University of Florida
Hong	Chuang	Virginia Tech University
Jarrels	Trey	Shreckhise Nursery
Jennings	Christina	Tennessee State University
Jones	Jeff	Circle J Tree Farm
Jones	Stacy	NC State Cooperative Extension
Kanosky	Jeff	Utopian Plants
Kopas	Nicole	Virginia Department of Agriculture
LaForest	Joseph	SIPM/University of Georgia
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Locke	Todd	BWI
Lookabaugh	Emma	BASF

Lynn	James	SiteOne Landscape Supply
Marshall	Sam	North Carolina State Extension
Martin	Michael	AmericanHort/HRI
Mattice	Stephen	Colesville Nursery
McCarter Jr.	Tom	OHP
McCulloch	Bryan	Mountain Shadow Nursery
Monday	Jeff	SiteOne Landscape Supply
Moore	Benjamin	USDA-ARS and Moore Nursery
Morrison	Charles	Tennessee State University
Munster	Mike	North Carolina State Univ. Extension
Neubauer	Alex	Hidden Hollow Nursery
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Parajuli	Madhav	USDA-ARS
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Searer-Jones	Kristen	IR-4/University of Florida
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Velasquez	Jared	Colesville Nursery
Vinatzer	Boris	Virginia Tech University
White	Elizabeth	Purdue University
Wilkerson	Carsen	Rainfrost Nursery
Weiland	Jerry	USDA-ARS
Witcher	Anthony	Tennessee State University
Yang	Xiao	Clemson University
Zampini	Maria	Proven Winners