Progress in the National Clean Plant Network







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NCPN Coordinator
USDA-APHIS-PPQ

Goals for this session

- Highlight the program progress and technical advances of the National Clean Plant Network over the program history
- Discuss cross-network areas of focus and future strategies to sustain into the future

What is the National Clean Plant Network?

- An association of clean plant centers, scientists, educators, state and federal regulators, nurseries, and growers of specialty crops that work together to ensure the availability of healthy plant propagation material.
- Clean plant centers produce and distribute disease-tested propagative plant material to protect specialty crop producers and the environment from the spread of harmful diseases.

Start clean, stay clean

Program History

- Industry, university, state regulatory, and USDA met in 2006-7; created a steering committee to develop the NCPN concept
- 2008 Farm Bill established NCPN as “a network of clean plant centers for diagnostic and pathogen elimination services to (1) produce clean propagative plant material; and (2) maintain blocks of pathogen-tested plant material in sites located throughout the United States.”
- 2009 - NCPN Federal partners (APHIS, ARS, NIFA) established Memorandum of Understanding and Governing Board
- Reauthorized and made permanent with 2014 and 2019 Farm Bill with at least $5 million in annual funding, and incorporated into the Plant Protection Act Section 7721

The need for clean plant material

- Many specialty crops are propagated vegetatively rather than through seed
- Viruses, and other internal pathogens, can be difficult to detect and can be inadvertently passed on to progeny and brought to the field
- Diseases that can be spread through infected plant material such as grapevine leafroll disease, little cherry disease, and citrus greening can cause devastating economic losses
- Starting new plantings with clean stock is the first step in ensuring a healthy, productive crop.

Network Crop Groups

- 2009 Grape and Fruit Tree
- 2010 Citrus, Berries, Hops
- 2015 Rose and Sweet Potato

A Collaborative Network

NCPN Management	Governing Board	Crop Governing Bodies	Clean Plant Centers	Working Groups
National networking and coordination Manages funding mechanism	Forum for critical discussions Advises on funding decisions Federal and state regulatory members	Includes research, extension, regulatory, and industry members Manages charter, membership, prioritization Planning, collaboration, advocacy for crop needs	Conducts diagnostics & therapeutics, establishes & manages foundation materials Delivers services and plant material to industry Recipient of national funds	Strategic Planning Education and Outreach Economics Quality

Network Program Funding

Program funding

- $8 million in FY2025, administered by USDA-APHIS as part of PPA 7721 program
- From 2009-2025, APHIS has provided close to $96 million for projects in 19 states and Puerto Rico.

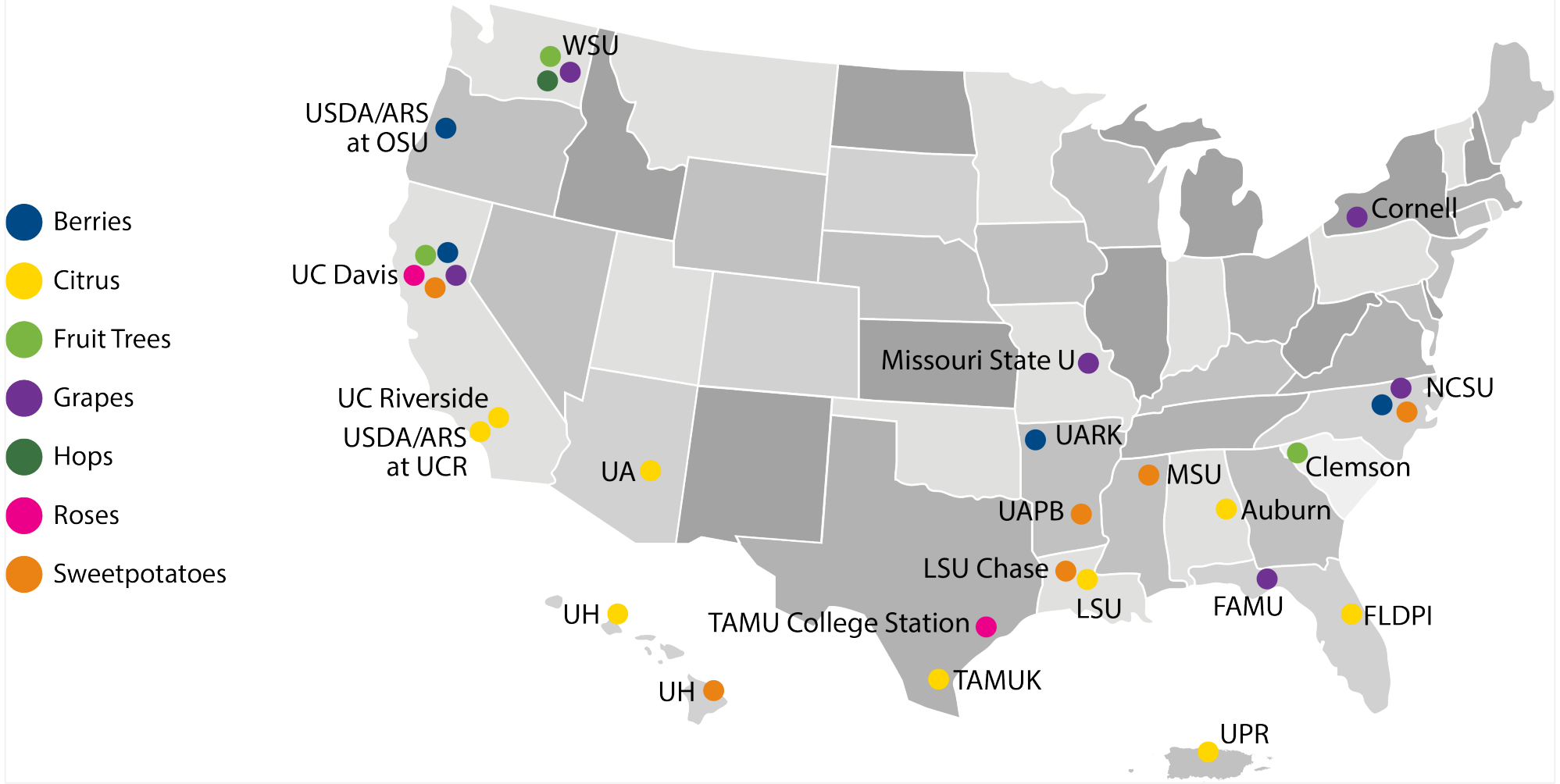
NCPN funds support

- Critical staffing needs
- Equipment and supplies
- Diagnostics, therapeutics, foundations
- Network governance and special initiatives

Other funding

- Centers also receive support from diverse sources including user and service fees, and commodity programs, as well as staff and infrastructure support from their institution

National Clean Plant Network Centers



Features of the Network Model

- Distributed model of service with partial financial support from federal funds. Clean plant programs are hosted by universities and state and federal agencies.
- Diverse size and scope for centers from national, multicrop focus to regional, single crop.
- Brings centers, industry, research, state and federal regulatory participants together to collaborate.
- Most center directors are also involved in research or extension. Directors may provide subject matter expertise on plant diseases, diagnostics, and regulatory issues, and conduct research that is complementary to NCPN.

The Clean Plant Process

Receipt of new accession from domestic or foreign sources

Plant material tested for pathogens

Tests negative for pathogens

Plant tested again for pathogens (and tests negative)

Tests positive for pathogens

Virus elimination attempted using methods such as tissue culture, heat therapy, or cryotherapy.

Propagation of new plants

Plant is released by Clean Plant Center or entered into foundation collection

Distribution of plant material to users

Center conducts regular maintenance testing of foundation collection to ensure plants remain free of pathogens of concern

Diagnostics

Therapeutics

Distribution

Foundation

NCPN and Role in Imported Germplasm

- Depending on type of crop and previous testing, varieties may be required to be imported by controlled import permit into quarantine. Material must test free of damaging pathogens before release.
- 9 NCPN centers have plant quarantine permits for citrus, grapes, pome and stone fruit, hops, berries, and rose
- Major component of U.S. capacity to safely introduce new varieties.
- NCPN centers can also provide APHIS-approved diagnostic testing for companies with quarantine facilities.

Overall Impacts of the National Clean Plant Network

Annual Deliverables

- Maintain over **8000** clean plant accessions
- Distribute about **1.5 million** propagative units for further increase
- Release **~450 new clean varieties** to industry from domestic and international sources

Market Protected

- Specialty crop industries served by NCPN have a collective annual value of production of **over $20 billion** and account for over **3.2 million acres** of crop production.

Return on Investment

- A single clean plant can be the source of thousands or even millions of field plants through additional propagation by nurseries.
- Clean plant programs provide economic returns of **up to 100-400** times the program costs and **billions in benefits** to growers by reducing crop losses to diseases.

Crop Services and Impacts

Citrus

- Foundation: over 2800
- Distribute: over 500,000 units of clean budwood and seed annually

- Almost all commercial citrus is derived from NCPN sources

Grapes

- Foundation: over 2600
- Distribute: over 35,000 units of clean grape material annually

- Provide grape material for at least 32 states

Fruit Trees

- Foundation: over 850
- Distribute: over 35,000 buds and 145,000 seeds annually

- NCPN centers are the source of clean material for trees such as apple, pear, cherry, peach, plum, and olive.

Berries

- Foundation: over 400 berry accessions, for blueberry, strawberry, raspberry, and blackberry; one clean plant can be expanded to millions in a few years
- Nursery sales of 800 million plants in CA and 200 million plants in FL produced annually from clean sources

Hops

- Foundation: ~50 hop accessions; each propagative unit can be expanded to thousands rapidly.
- Supporting about 30% of the world's need for clean hops.

Sweet Potato

- Foundation: ~400 clean sweet potato accessions
- Distribute: about 560,000 units of seed, slips, plants, and tissue culture plants annually in six states

Rose

- Foundation: ~1000 rose accession
- Distribute over 25,000 clean cuttings annually to nurseries.
- The rose industry produces about 30 million plants and contributes $534 to $777 million in direct economic impacts to the US annually.

NCPN Progress and Advances

- New diagnostics methods validated by network centers, such as high throughput sequencing, can save 1-3 years in the process for new imported varieties.
- Advances in tissue culture and therapy treatments have increased the speed and effectiveness of production of clean plants
- Information management solutions provide access to plant tracking and testing results directly to customers
- Germplasm virology and diagnostics expertise supports regulatory updates

Benefits of NCPN to Stakeholders

- Increased support to clean plant centers increases availability of clean plants
- Nurseries have access to clean propagative material, improving the quality and value of the plants they sell
- Growers have economic benefits from higher crop yield and quality, and reduced losses due to disease
- Industry has increased access to new varieties while preventing risk of disease spread
- Establishes a network of cooperating scientists and stakeholders and forum for discussion and collaboration
- Provides linkages to state and federal plant protection regulatory programs

Learn more at: www.nationalcleanplantnetwork.org






NCPN Advances at Foundation Plant Services

September 10, 2025

Maher Al Rwahnih

UCDAVIS
Foundation Plant Services

Current Plant Collections

Grapevine
2,504 selections

Rose
1,009 selections

Strawberry
34 selections

Sweetpotato
64 selections

Fruit Trees

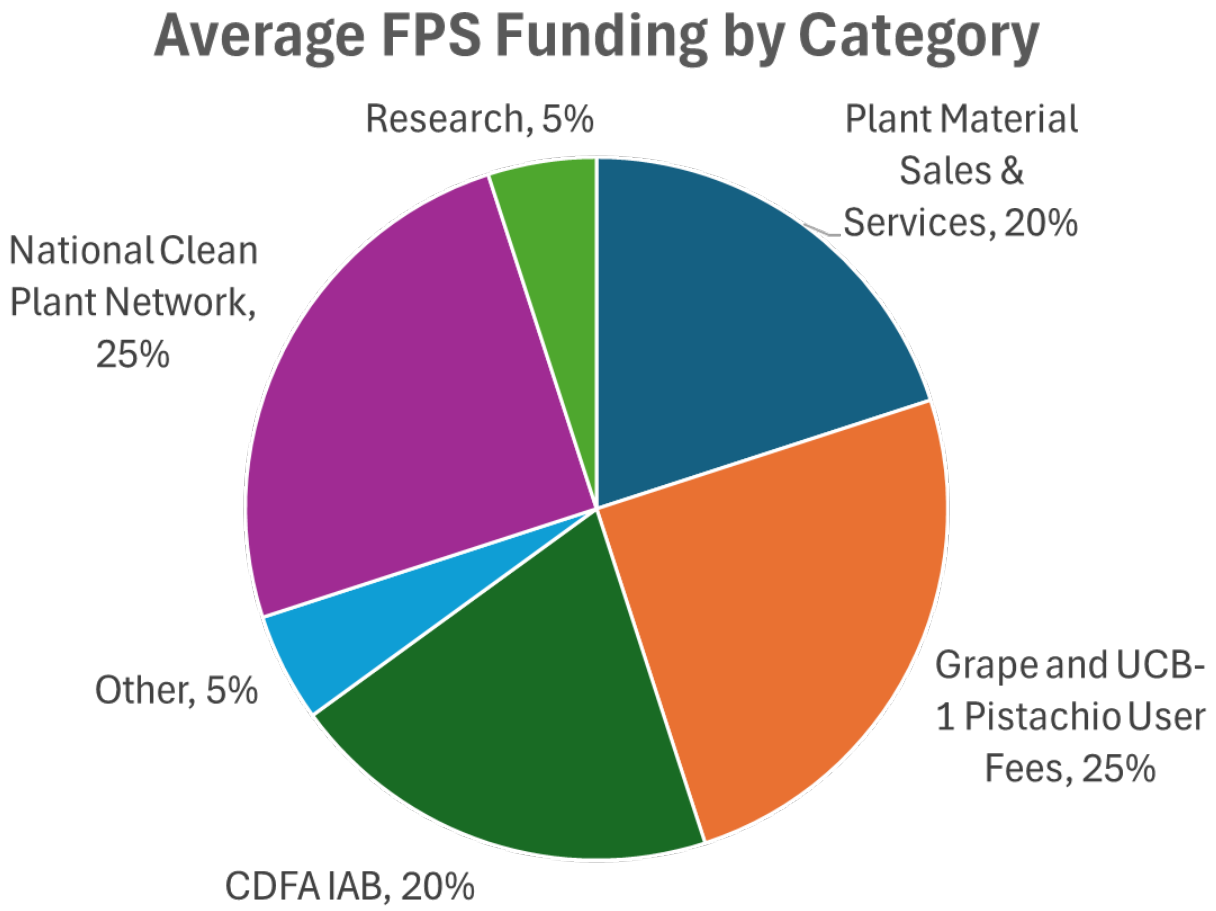
Prunus, Pyrus, Malus
603 selections

Olive
39 selections

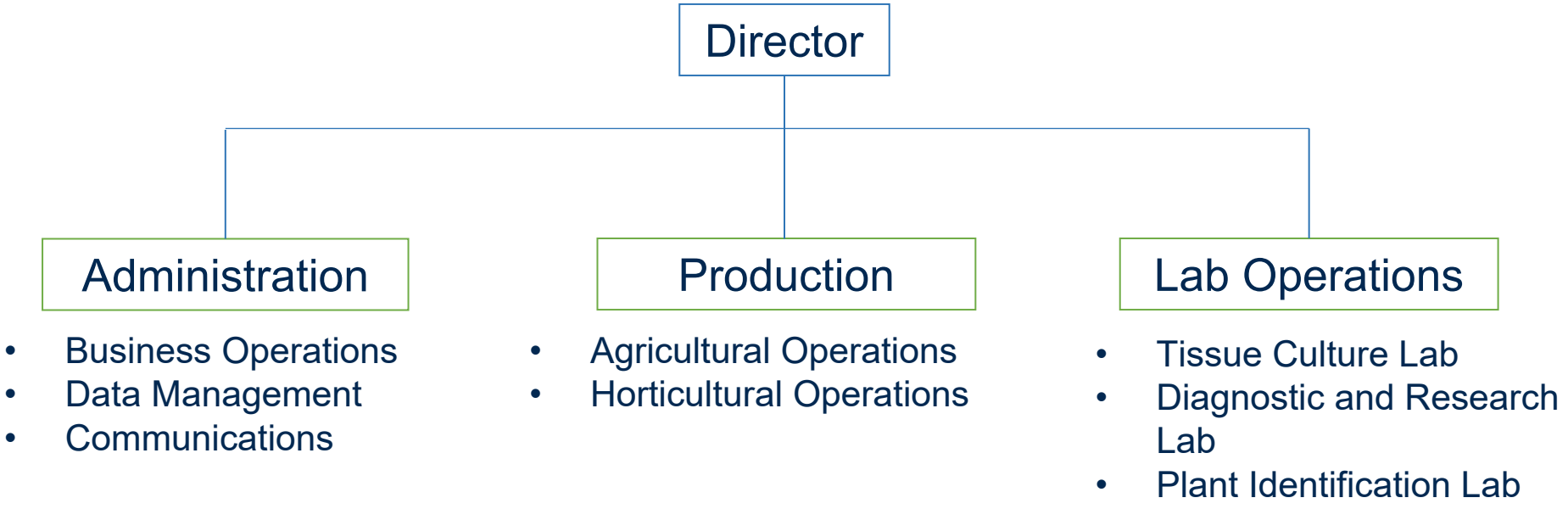
Counts as of August 2025

NCPN Funding to FPS

- NCPN provides about 25% of FPS funding each year
  - Important for operations of the clean plant program



FPS Organizational Structure



- Business Operations
- Data Management
- Communications

- Agricultural Operations
- Horticultural Operations

- Tissue Culture Lab
- Diagnostic and Research Lab
- Plant Identification Lab

- **Self-supporting** unit in the UC Davis College of Agricultural & Environmental Sciences
- ~45 staff
- **Annual budget $7-7.5 million**

Distribution of Clean Material

Each year, we distribute on average:

- 30,000+ grape cuttings and plants
- 30,000+ fruit tree buds and cuttings
- 240,000+ fruit tree rootstock seeds
- 80,000+ rose cuttings
- 15,000+ sweetpotato cuttings and rooted plugs

Foundation Collection Maintenance

- NCPN funding allows us to maintain these source clean plant protections
  - Pruning & other cultural work
  - Insecticide and herbicide treatments
  - Annual testing to confirm virus-free status

Annual Testing Panel – Outdoor Vineyard
Grapevine leafroll-associated virus 1
Grapevine leafroll-associated virus 2
Grapevine leafroll-associated virus 2 Redglobe strain
Grapevine leaf roll-associated virus 3
Grapevine leaf roll-associated virus 4
Grapevine leaf roll-associated virus 4 strain 5
Grapevine leaf roll-associated virus 4 strain 6
Grapevine leaf roll-associated virus 4 strain 9
Grapevine leafroll-associated virus 4 strain Pr
Grapevine leafroll-associated virus 4 strain Carnelian
Grapevine leaf roll-associated virus 7
Grapevine virus A
Grapevine virus B
Phytoplasmas (universal)
Tomato ringspot virus
Xylella fastidiosa
Grapevine red blotch virus
Grapevine fanleaf virus
Grapevine fleck virus
Grapevine Pinot gris virus

Annual Testing Panel – Orchard
Cherry virus A
Little cherry virus 1
Little cherry virus 2
Prunus necrotic ringspot virus
Prune dwarf virus
Plum pox virus
Phytoplasma Western X
Phytoplasmas (universal)

Annual Testing Panel – Sweetpotato
Ipomoea setosa index
Sweet potato feather mottle virus
Sweet potato leaf curl virus
Sweet potato chlorotic stunt virus
Ipomoea vein mosaic
Sweetpotato virus G
Sweet potato virus C

NCPN-Sweetpotatoes at FPS

- 64 selections
- Most material is distributed to California producers
  - Distribute rooted and unrooted cuttings.
  - Recent economic evaluation by Scott Stoddard reinforces it is important to use clean stock!
- The program is operating at a deficit
  - Feel it is important to provide services for our regional growers.
  - Evaluating collection composition and management

NCPN-Roses at FPS

- FPS hosts the only Rose collection in the Network.
- 1,008 cultivars

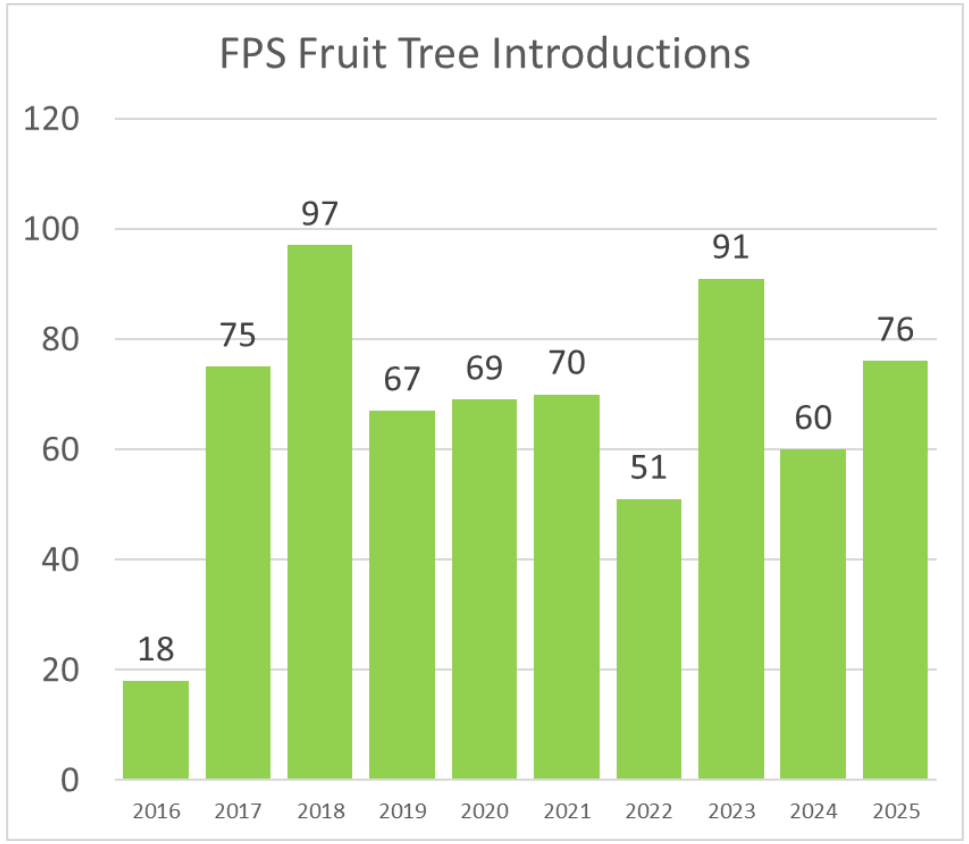
- Rose program has been operating at a deficit for 10+ years.
- We must change how we are doing things
  - Rose Tier 2 board is very supportive of two-pronged approach:
    - 1) Increase fees and charge for services that were previously no-cost.
    - 2) Review cultivars in collection, and cull those that industry does not want.

NCPN-FT at FPS

- More cultivars added to Fruit Tree Foundation each year
  - 603 selections
- NCPN-FT now includes olives
  - Crop gaining more acres in California
  - 39 olive selections now at FPS
- Foundation Apple and Pear blocks added in 2023.

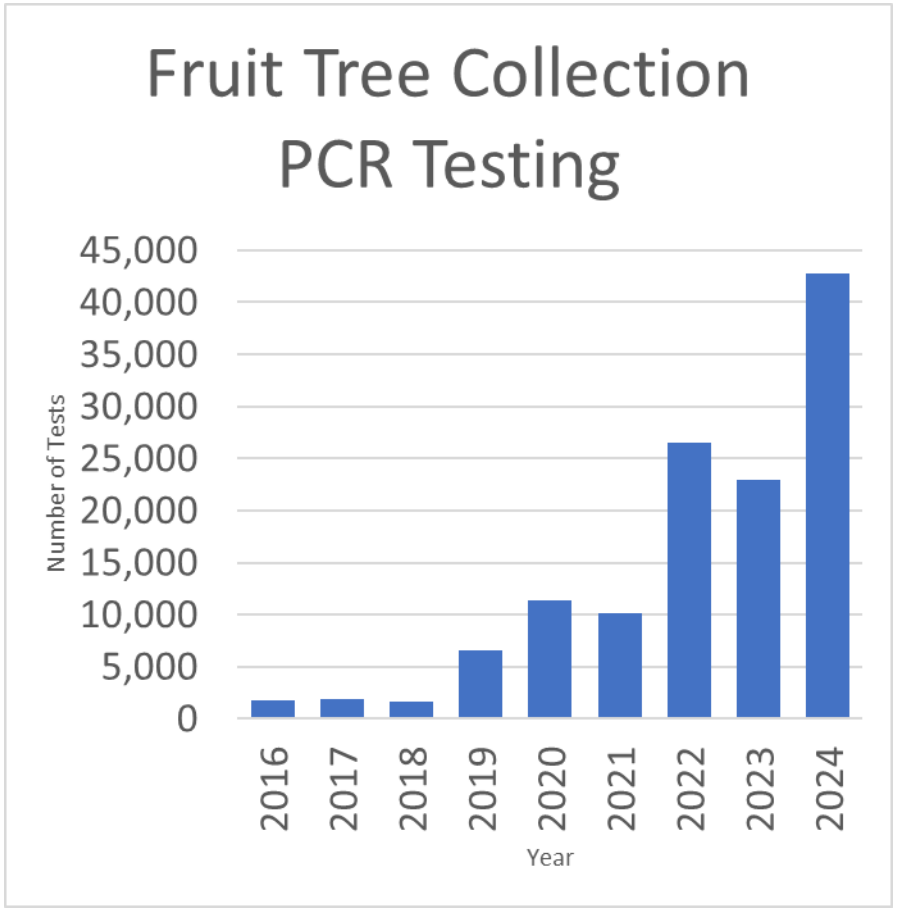
Fruit Tree Program Growth

- FPS has maintained *Prunus* material since its establishment in the 1950s, because of virus issues and crop loss.
- Grape was the main FPS import program, Fruit Trees added in 2017
- Now, demand for Fruit Tree services often exceeds grapes.



Fruit Tree Program Growth

- Testing in diagnostic lab has dramatically increased as the program grows
- PCR used to screen introductions
- PCR used on annual testing
  - LChV-1, LChV-2, phytoplasmas added to testing panel due to little cherry disease out break in PNW.
  - CVA added due to risk of pollen transmission.



FPS Grapevine Collection

Classic Foundation Vineyard
2,504 selections

Foundation Greenhouse
~1,200 selections
Most, duplicating Classic material

Propagation Work Continues

- With 2,500 total selections in the collection, we have more to propagate and protect!
- NCPN operational support has contributed to the propagation work to move grapes indoors.
- **The greenhouse is full** , so we are making more space.

Coming Soon: Foundation Screenhouse

- 12,000 square feet
- Fully screened for insect exclusion
- $2.7 million bid for construction
  - $2.5 million loan from College allowed us to start the project.
- Construction team on site September 1.
- Expected completion April 2026

Classic Foundation Vineyard Testing History

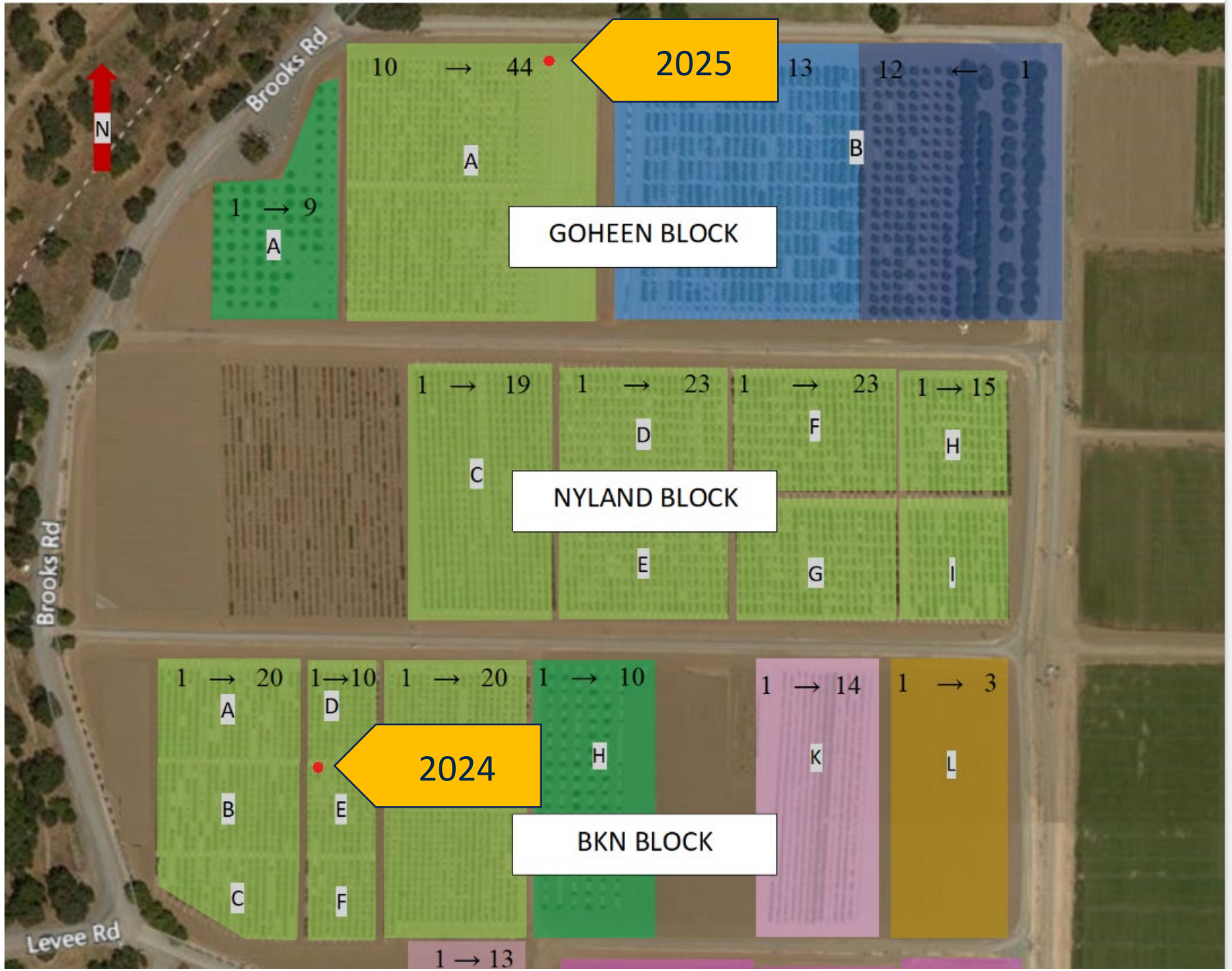
Year	Vines Tested	Total Vines*	GRBV Positive Vines	GRBV Incidence	GLRaV-3 Positive Vines	GLRaV-3 Incidence
2017	3,604	4,088	1	0.02%	0	0%
2018	4,127	4,075	0	0%	0	0%
2019	4,167	4,075	1	0.02%	0	0%
2020	4,301	4,270	0	0%	0	0%
2021	4,594	4,594	0	0%	0	0%
2022	4,660	4,660	0	0%	0	0%
2023	4,634	4,634	0	0%	0	0%
2024	4,881	4,881	0	0%	1	0.02%
2025	4,991	4,991	1	0.02%	1	0.02%

Classic Foundation Vineyard – 2025 Virus Testing

- Of 4,991 vines at Classic Foundation Vineyard:
  - 1 positive for GLRaV-3
  - 1 positive for GRBV

  = 2 vines removed in August, immediately upon virus detection

Location of GLRaV-3 Positive Vines



UCDAVIS
Foundation Plant Services

Urgency to Propagate for Indoors

- The screenhouse will be ready by spring 2026
- LR3 and GRBV detections at Classic Vineyard are a problem!
- CDFA Pierce's Disease Control Board provided propagation-specific funding to support up to 50 selections for 2025-2026.
- Seeking propagation funding from other sources so work can continue at full capacity.

Operational Challenges

- California Nursery Industry is struggling with reduced sales
- This trickles down to FPS as reduced sales, nurseries generating fewer user fees, and reduced funding available from CDFA and industry groups.
- In response to reduced operational funding, we are:
  - Increasing automation where possible
  - Cutting back expenditures on supplies, maintenance.
  - Reducing staff: 3 layoffs this year
  - Working across departments on special projects

Above: Diagnostic lab staff assist with pruning Classic Foundation Vineyard

Import & Quarantine Program

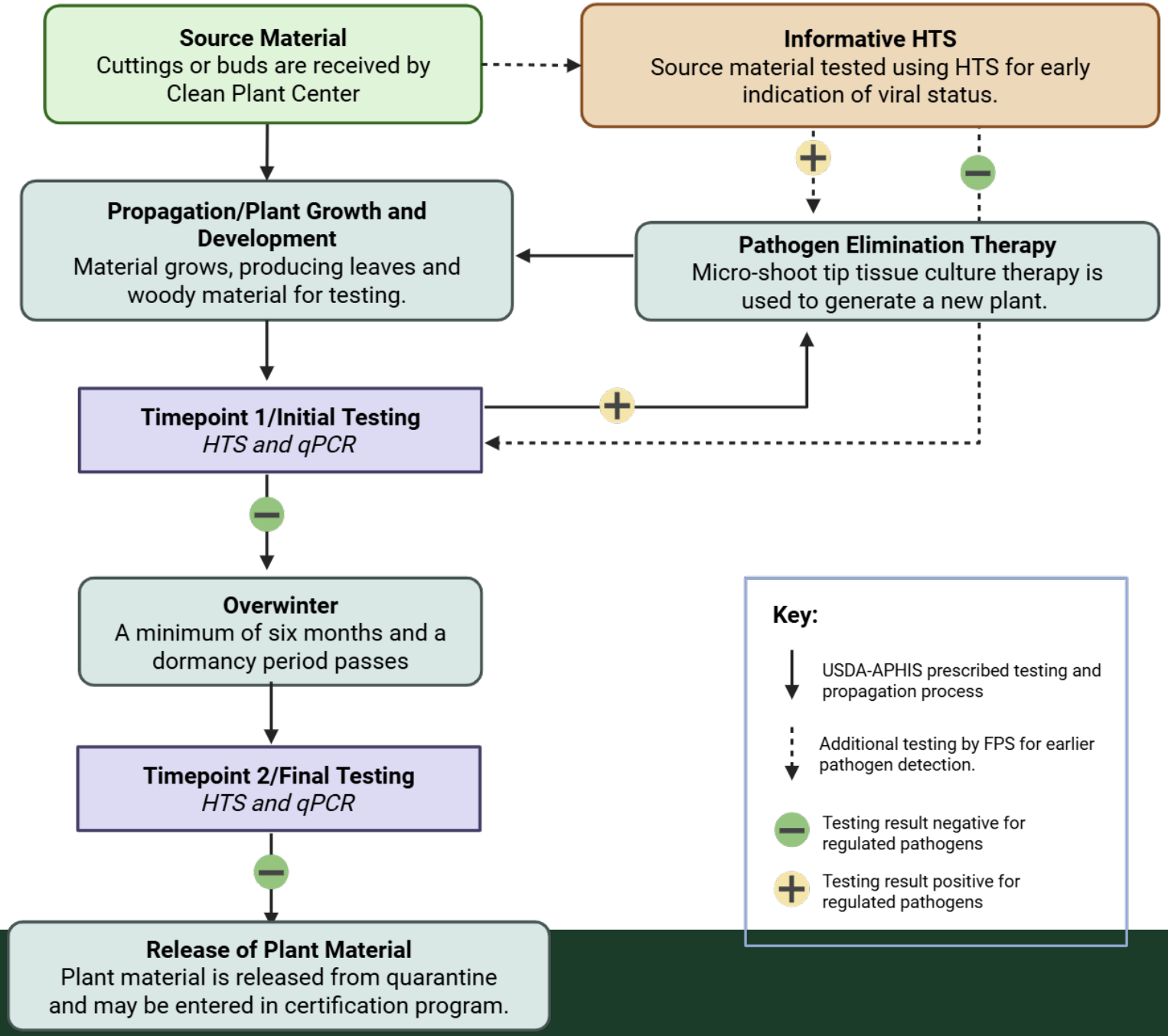
Import & Quarantine Program

- 588 Controlled Import Permit allows import of:
  - Grapes
  - Fruit Trees (*Prunus, Malus, Olea, Pyrus*)
  - Berries (*Rubus, Vaccinium, Fragaria*) *NEW*
  - Rose
- We use High Throughput Sequencing to screen quarantine material for viruses.

Import & Quarantine Programs

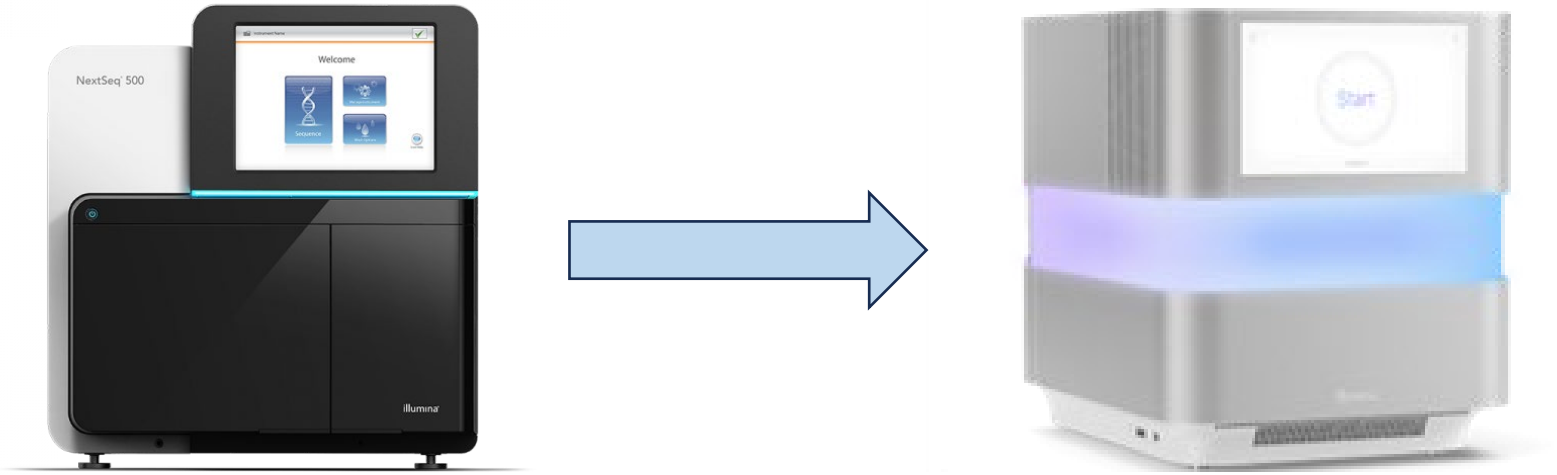
- High demand for import & quarantine, testing & treatment
- Average 70 Grape introductions each year
  - 40 from international sources, 30 domestic sources
- Average 70 Fruit Tree introductions each year
  - 50 from international sources, 20 domestic sources
- Rose program averages 6-10 introductions each year
- Berries – new import program at FPS as of 2025, many requests, and just starting to receive shipments.

Testing with HTS



Building HTS Capabilities

- 2007: FPS started using HTS
- 2016: Purchased NextSeq 500 (Illumina)
  - Equipment partially funded by NCPN (~$100,000)
- 2024: Upgraded to NextSeq 2000 (Illumina)



NCPN Investments that Launched HTS:

Inter-laboratory validation and standardization of high throughput sequencing (HTS) protocol for detection of regulated pathogens for specialty (nursery) crops

- Funded by NCPN/PPA 7721

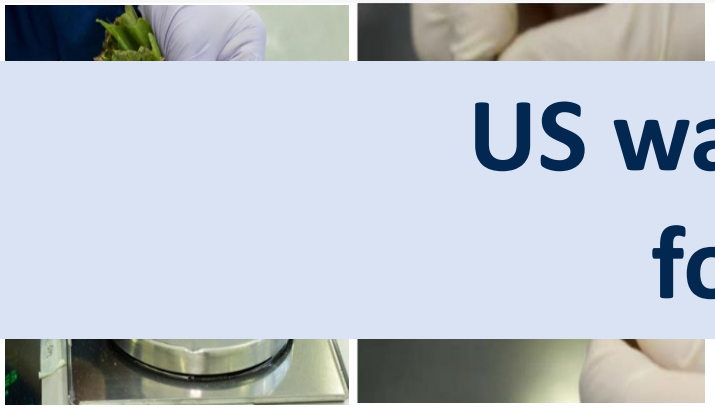
Objectives:

- *1.* Validation of TruSeq Total RNA, Ribo-depleted protocol for HTS of selected specialty crops (Apple and Grapevine)
- *2.* Validation of HTS-Bioinformatic pipelines used by participating laboratories for the identification of plant viruses and virus-like agents in specialty crops
- *3.* Development of standard operating procedures (SOPs) and minimum guidelines for harmonization and implementation of HTS on other specialty crops.

Revised Grapevine and Fruit Tree SOPs

HTS analysis

RT-qPCR/PCR



US was first country to adopt HTS for testing regulated material

Test two different types of plant material at two time points

X No biological indexing

Approved by CDFA and USDA APHIS

Success Story Subject of Feature Article

Plant Disease • 2025 • 109:1392-1403 • https://doi.org/10.1094/PDIS-10-24-2104-FE

PLANT DISEASE FEATURE

A New Era in Federal Quarantine and State Certification Diagnostics at Clean Plant Centers in the United States

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Funding: Support was provided by National Clean Plant Network and California Department of Food and Agriculture Improvement Advisory Board.

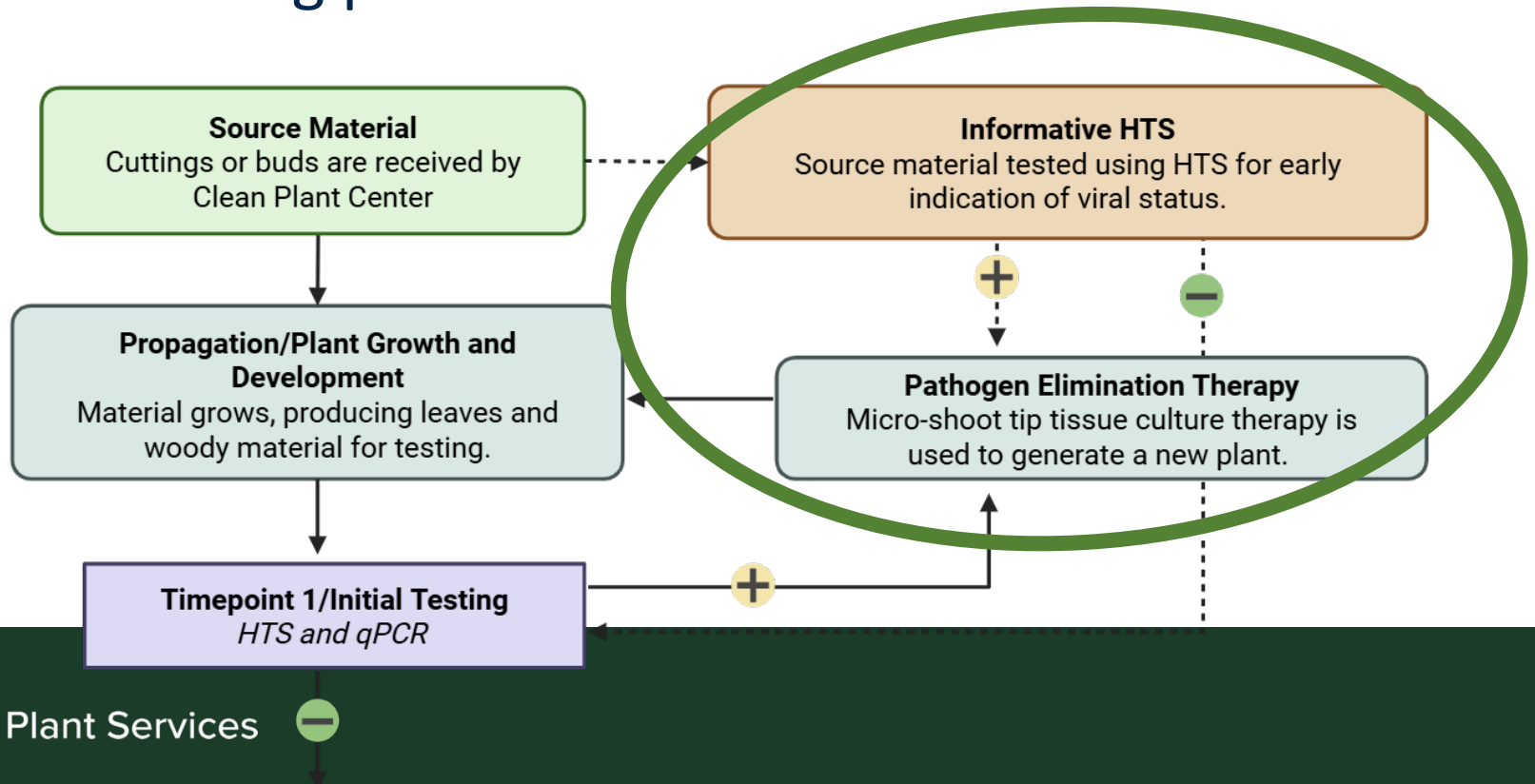
Abstract

NCPN Impacts in the Diagnostic and Therapeutic Work at FPS

HTS: More Results in Less Time

Woody indexing to determine disease status could take **2-3 years**.

- Now, we get our first determination of virus status within weeks or months of receiving plant material.



HTS: Early Indication of Viral Status

Using HTS results, we decide if virus elimination therapy is needed.

- Previously: all grapevine introductions went through tissue culture before index results (virus status) was known.
- Of 50 grape intros in 2024, 25 require tissue culture therapy
  - The other 25 continue in the testing pipeline without requiring the cost or time of therapy.
    - Most will be released from quarantine based on summer 2025 test results.

HTS Shortening Quarantine

Indexing to determine disease status could take **2-3 years**.

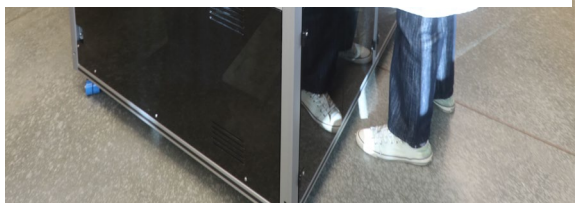
- Now:
  - Grapes, fruit trees can complete testing just **18 months** after arrival.

IMPACT: Clean material of important new varieties reaches growers in less time.

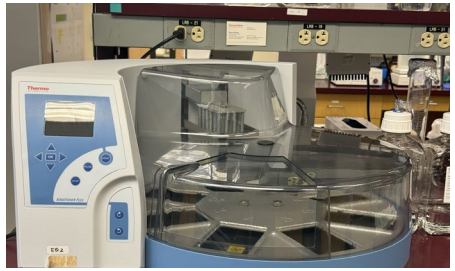
FPS DIAGNOSTIC LAB

- We have a well-equipped lab and experienced team and are able to do all required testing in-house.
  - This is key to maintaining the quality and accuracy of our results.

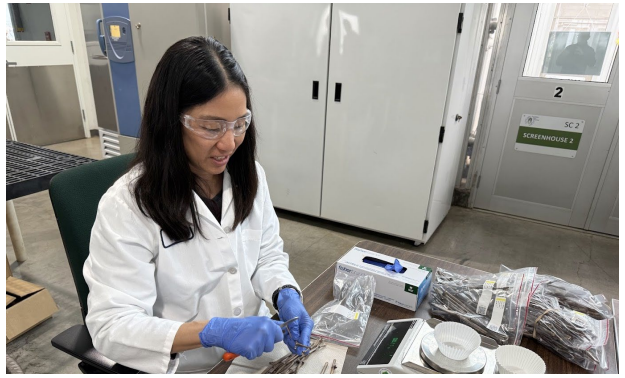
AUTOMATED LIQUID HANDLER



NEXTSEQ 2000 SEQUENCER



TNA EXTRACTION ROBOT



SAMPLE PROCESSING

BIOINFORMATICIAN

Ready and Able to Test for Others

- You do not need to build a lab like ours to conduct your HTS testing
  - Use our lab and team to test your samples!
- We are collaborating on research, but also on clean plants.
- Example in Grape NCPN centers:
  - For Cornell and Midwest Centers we have agreements to do their testing.
    - Can extend this service to others.

NEW: FPS NCPN Berries Program

- Strawberry, blueberry, raspberry, blackberry
- One shipment (3 selections) already received.
- Expecting 6 additional shipments, 18 more selections from New Zealand, Australia, Netherlands and China




Improvements in Tissue Culture Lab

Ejay Dehal hired July 2024
Director of Production & Quality Assurance

- 10+ years industry TC experience

TC lab expanded with new hoods, more space

- Space was needed due to increased program demand

Improvements in Tissue Culture Lab

Two technicians trained in cryotherapy techniques at USDA/ARS in Colorado

Early screening to remove plantlets where therapy was unsuccessful

- Negative plantlets advance to soil and will be tested on regular pipeline.

Traceability & Customer Service

Tissue Culture Lab is latest department to add QR codes to plant material tracking

QR codes are used on plants in the nursery and field

Distribution of Clean Material

Each year, we distribute on average:

- 30,000+ grape cuttings and plants
- 30,000+ fruit tree buds and cuttings
- 240,000+ fruit tree rootstock seeds
- 80,000+ rose cuttings
- 15,000+ sweetpotato cuttings and rooted plugs

Traceability & Customer Service

- QR codes are scanned using mobile devices
- When shipping material, the code inventories against packing list.
- In the field and nursery, this allows real time inventory and plant health records.

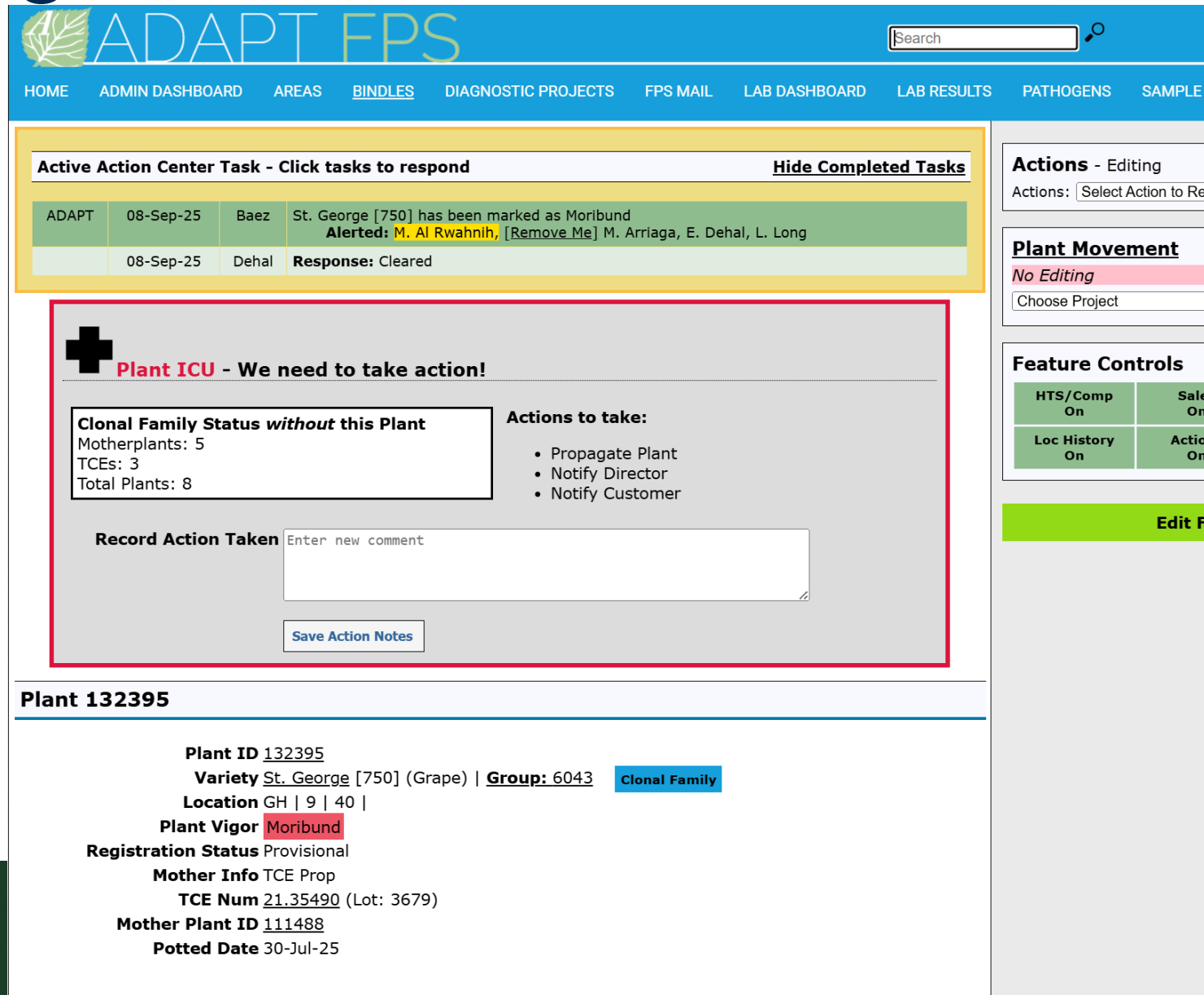
ADAPT Database Tool

- ADAPT was a tool built by FPS, for FPS.
- NCPN operational funding allowed us to direct plant sale monies to support the programming team.
- Programmers have lots of experience at FPS!
  - 11 years Karl, 4 years Andy
- This experience is now applied to 'ADAPT Core' database for NCPN
  - Karl will present later today.

Plant Health Management

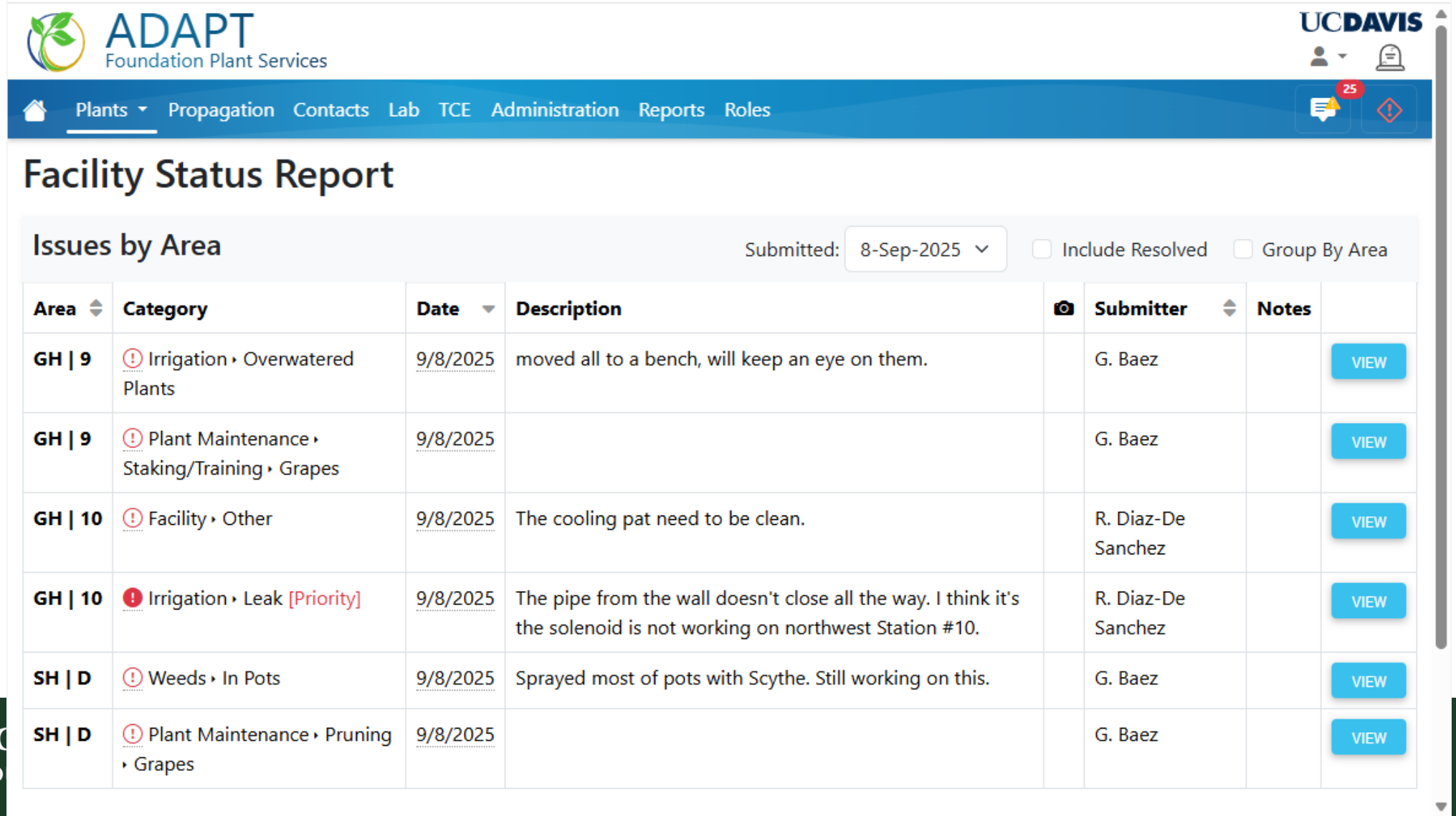
Example of tools in use:

- Nursery technician finds ailing plant.
- Scans QR code on pot to navigate to plant in database.
- Marks as Moribund
- Management is alerted to health status – to take action!



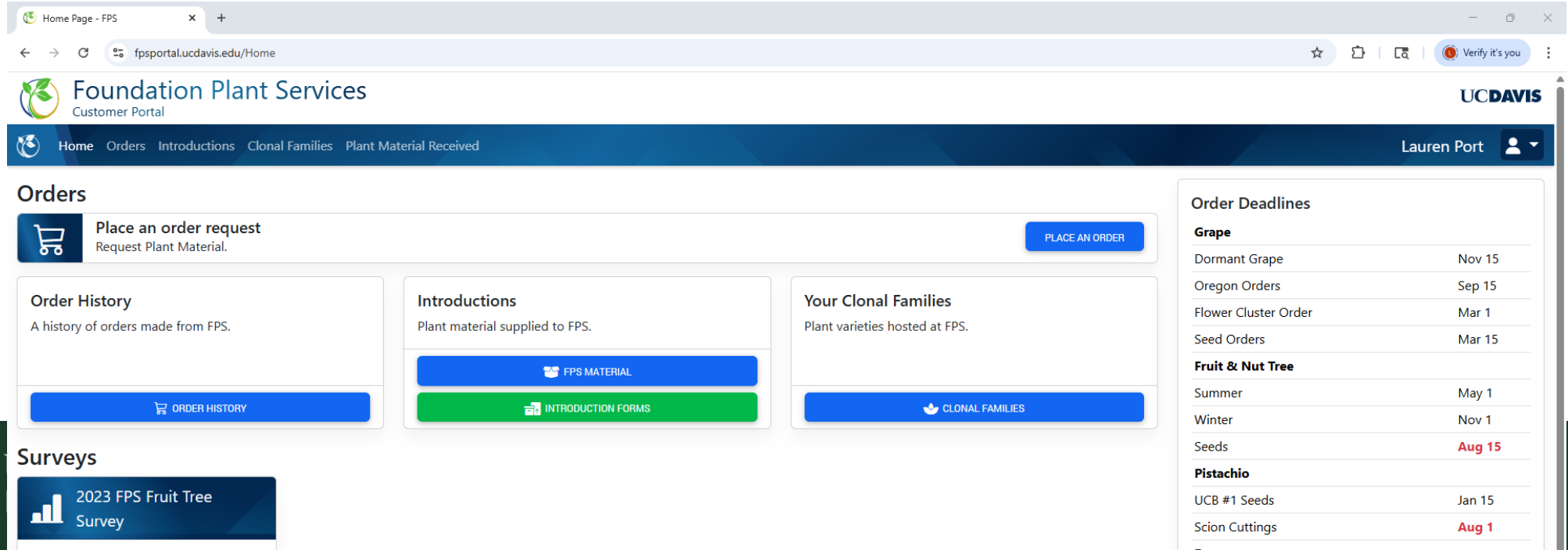
UCDAVIS Foundation Plant Services

Plant Health & Facility Maintenance



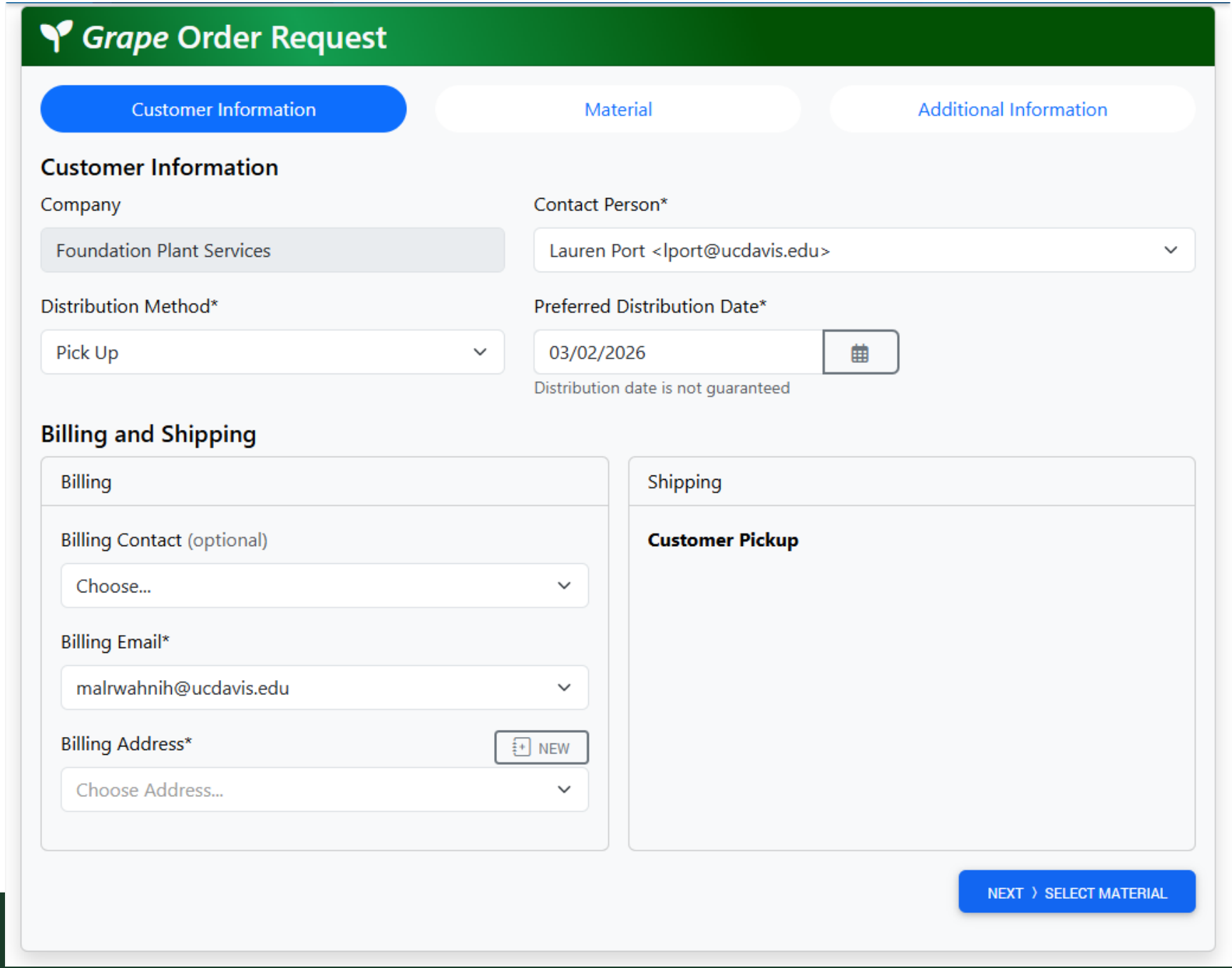
FPS Customer Portal

- 2024-2025 first year of use
- Forms in the portal are used for:
  - Order Submission
  - Introduction Requests



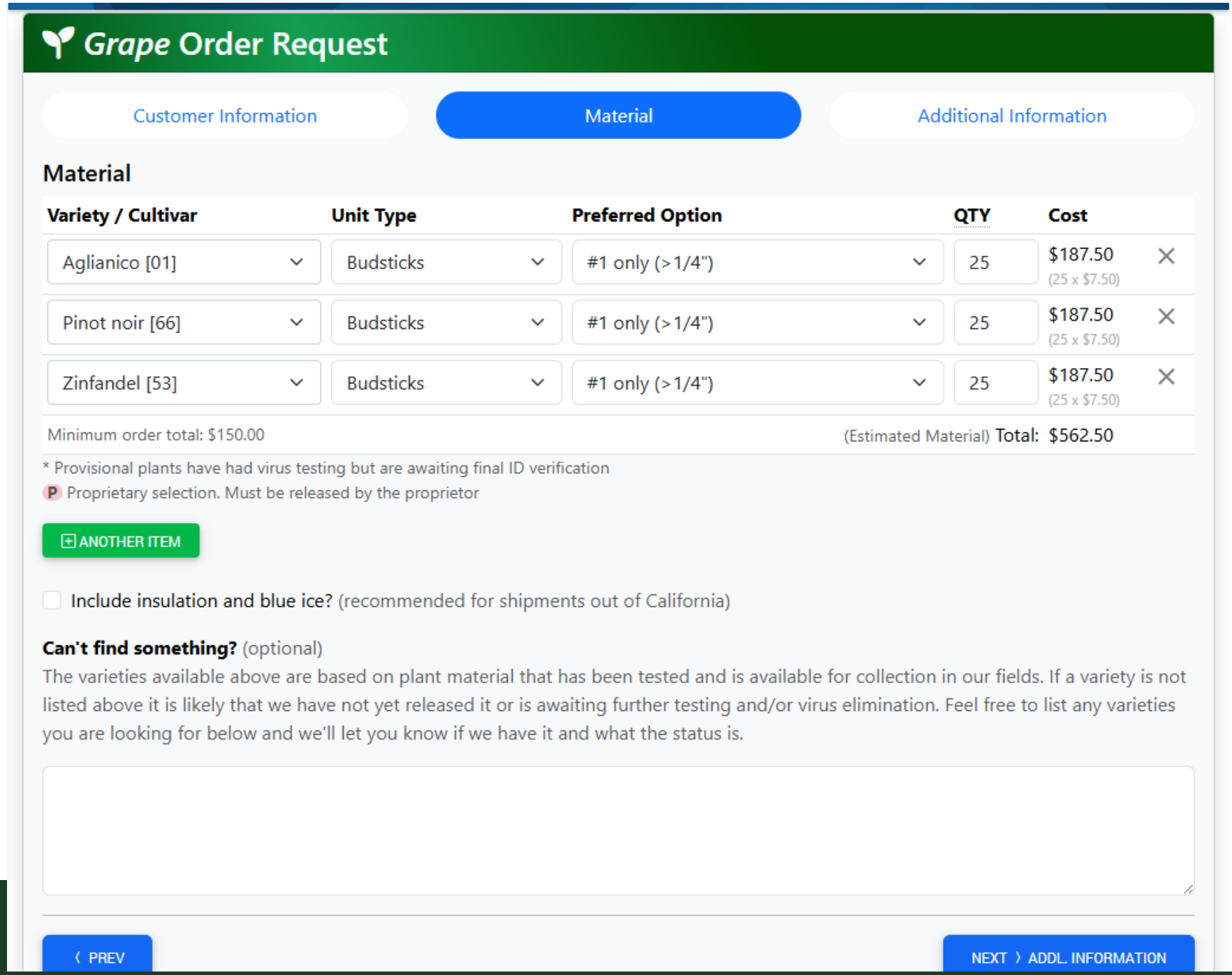
Order Entry

- No more interpreting handwriting
- Required fields must be completed = we get all the information we need!



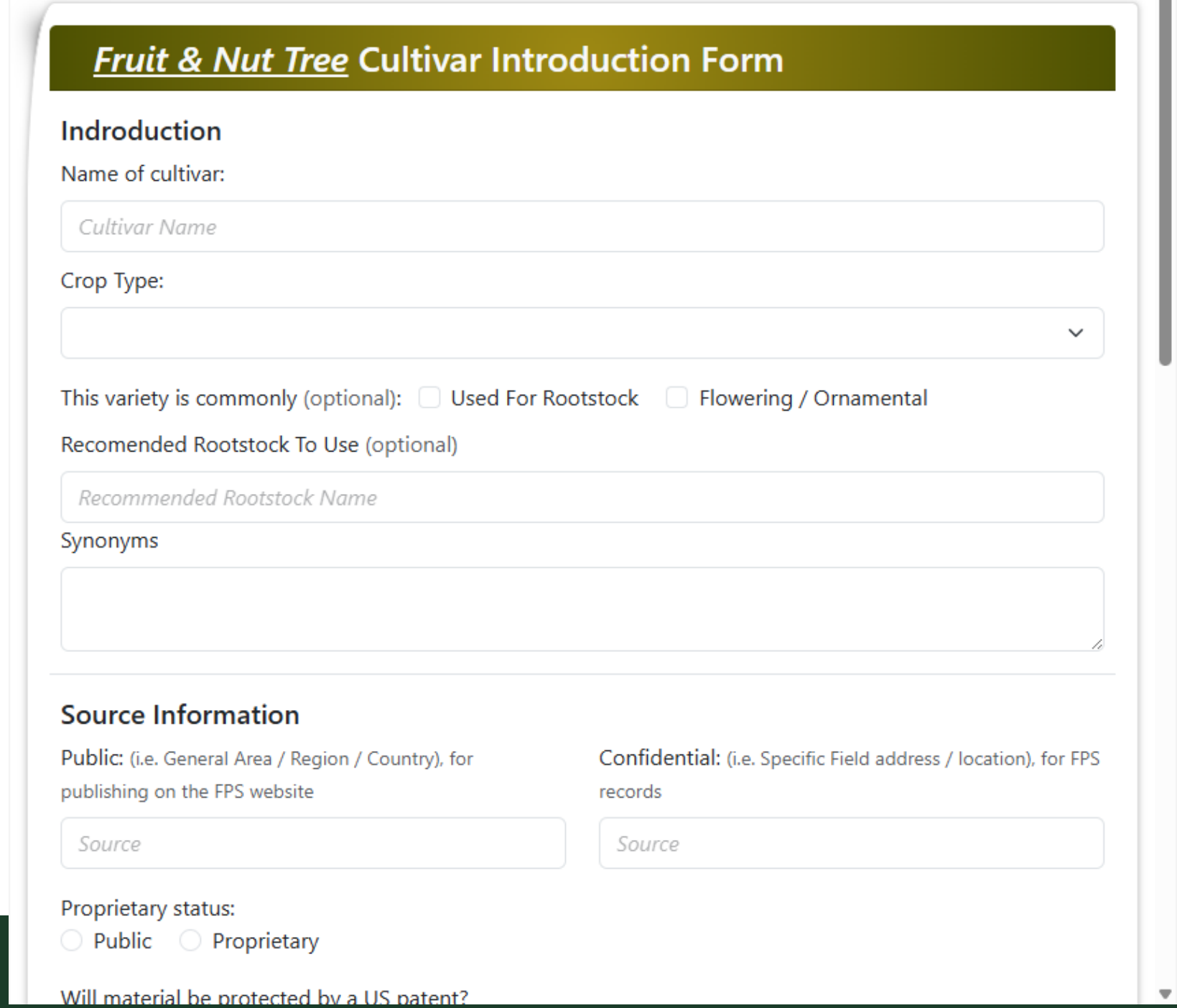
Order Entry

- Customers see exactly what selections are available
- Order policies are clear up front (i.e., $150 plant material minimum)



Introduction Forms

- Again, avoiding handwriting issues!
- FPS receives forms immediately after submission
- Documents link in ADAPT to import permit, labels, and shipment (after arrival)



Tools for Sponsors

- Shipment dashboard

Material supplied to FPS 6

Material supplied to FPS

Shipment #	Crop	Material	Country of Origin	Rece
2018-8	Grape	Uva Longanesi [01] (Grape) Albana [02] (Grape) Famoso [01] (Grape) Centesimino [01] (Grape) Cesanese [01] (Grape) Bombino Bianco [02] (Grape) Susumaniello [01] (Grape) Bombino Bianco [01] (Grape) Nero Di Troia [01] (Grape) Nerello Mascalese [03] (Grape) (Proprietary) Carricante [03] (Grape) (Proprietary) Glera [01] (Grape)	Italy	21-M
2018-3	Grape	Hondarribi Zuri [01] (Grape) Hondarribi Beltza [1] (Grape) Morenillo [01] (Grape) Sumoll [01] (Grape) Mandó [01] (Grape) Trepat [01] (Grape)	Spain	9-Feb
2017-13	Grape	Verdicchio [01] (Grape) Malvasia Istriana [01] (Grape) Verduzzo friulano [01] (Grape) Vitovska [01] (Grape) Albarossa [01] (Grape) Nascetta [01] (Grape) Pelaverga Piccolo [01] (Grape)	Italy	23-M

Tools for Sponsors

- Testing and release tracking

Clonal Families 36

Plant varieties hosted at FPS

Plant type	Material - Clonal Family	Proprietary	Contract Type	Release Date
Grape	**Glera [01]**	No	Introduction/Maintenance	1-May-2024
Grape	**Bombino Bianco [02]**	No	Introduction/Maintenance	29-Mar-2023
Grape	**Cesanese [01]**	No	Introduction/Maintenance	29-Mar-2023
Grape	**Mantonico [01]**	No	Introduction/Maintenance	29-Mar-2023
Grape	**Nerello Mascalese [02]**	No	Introduction/Maintenance	29-Mar-2023
Grape	**Albana [02]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Centesimino [01]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Famoso [01]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Mandó [01]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Sumoll [01]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Susumaniello [01]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Trepat [01]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Uva Longanesi [01]**	No	Introduction/Maintenance	4-Apr-2022
Grape	**Bombino Bianco [01]**	No	Introduction/Maintenance	23-Feb-2021
Grape	**Hondarribi Zuri [01]**	No	Introduction/Maintenance	23-Feb-2021
Grape	**Lacrima di Morro d'Alba [01]**	No	Introduction/Maintenance	23-Feb-2021
Grape	**Malvasia Istriana [01]**	No	Introduction/Maintenance	23-Feb-2021
Grape	**Morenillo [01]**	No	Introduction/Maintenance	23-Feb-2021

What's Next?

- FPS is handling multiple crops
- Can be support for other centers

- Oman, India, China
  - Great success story that other countries are emulating NCPN

Thank you, NCPN and FPS team!

Advances in Clean Citrus

WERA 20 - Raleigh, NC
NCPN Cooperators Session
September 10, 2025

Georgios Vidalakis, Ph.D.
Director, Citrus Clonal Protection Program
UC Presidential Researcher, Endowment for Sustainable Citrus Clonal Protection
Dept. of Microbiology & Plant Pathology
University of California, Riverside

Acknowledgments

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Fatima Osman
Stacey Comstock
Michelle Ortiz
Gerardo Uribe

TES Lab
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Ryan Milton

Texas
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Mark Van Ness

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Hailing Jin
Gregor Blaha
Mike Roose
Danelle Seymour
Tracy Kahn
James Borneman
Phillip Rolshausen
Caroline Roper
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Kim Bowman
James Thomson
Matt Matia

Brazil
Silvio Lopes
Juliana Astua
Nelson Wulff
Juliano Ayres

Australia
Nerida Donovan
Grant Chambers

South Africa
Glynnis Cook
Paul Fourie
Hano Maree

Chile
Juan Enrique Ortuzar
Ximena Besoain

EU
Leandro Pena

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Jude Grosser

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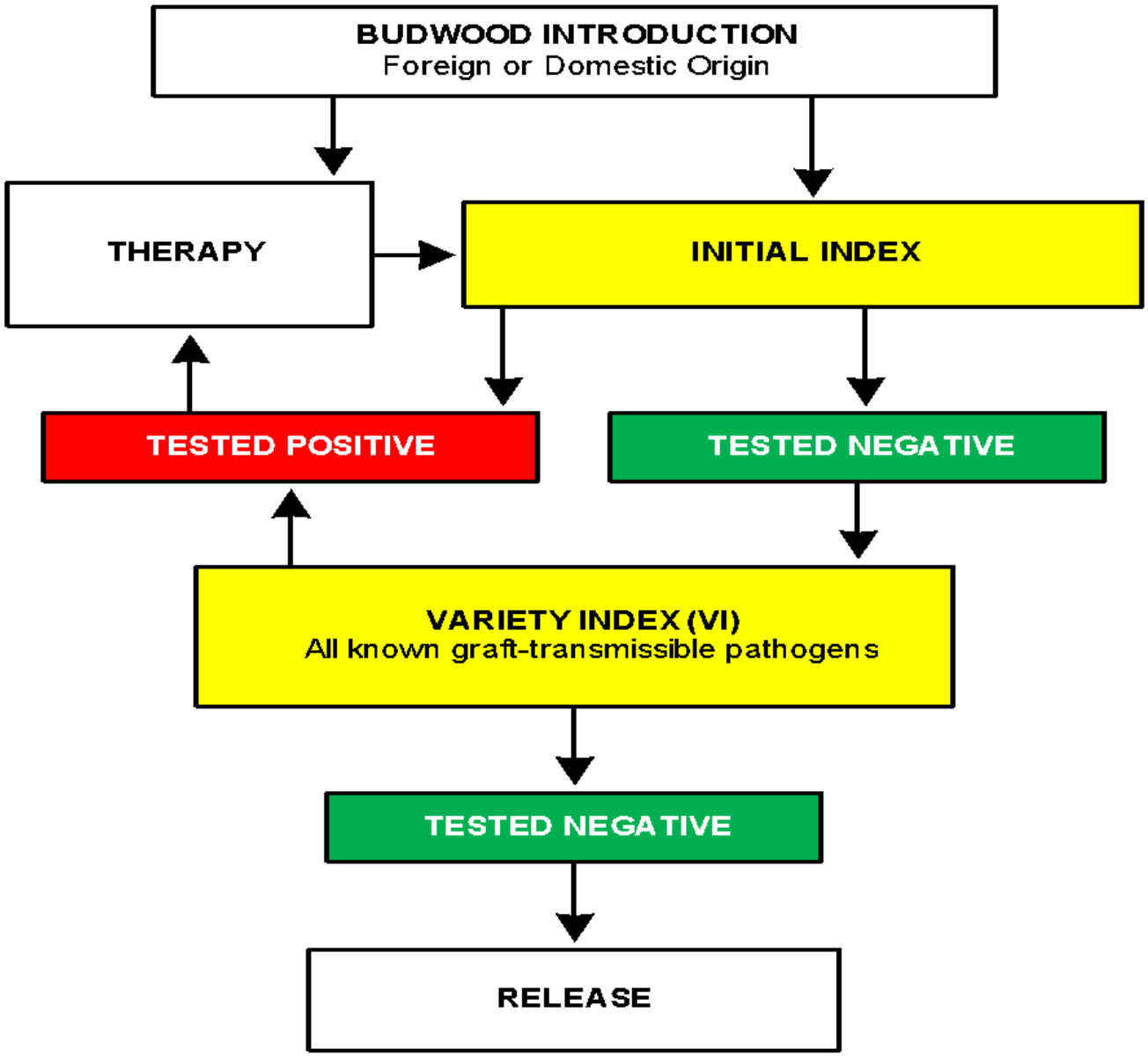
Justin Ezell
Maria Brenelli

NCPN
Jennifer Nicholson

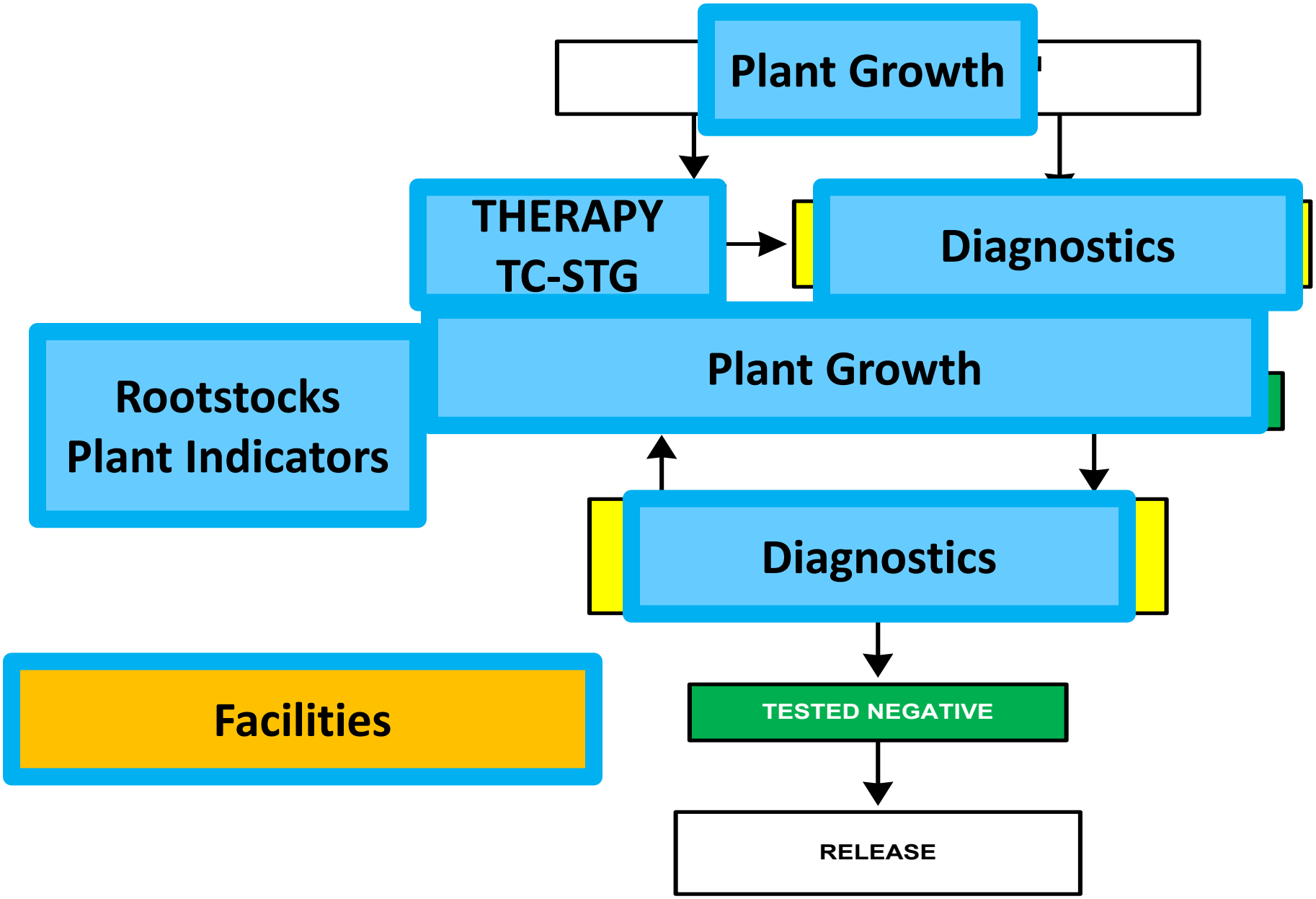
CDFA
Lucita Kumagai
Victoria Hornbaker

US Sugar-Southern Gardens
Mike Irey

Sacramento State
Jonathan Kaplan


BUDWOOD INTRODUCTION
Foreign or Domestic Origin
THERAPY
INITIAL INDEX
TESTED POSITIVE
TESTED NEGATIVE
VARIETY INDEX (VI)
All known graft-transmissible pathogens
TESTED NEGATIVE
RELEASE

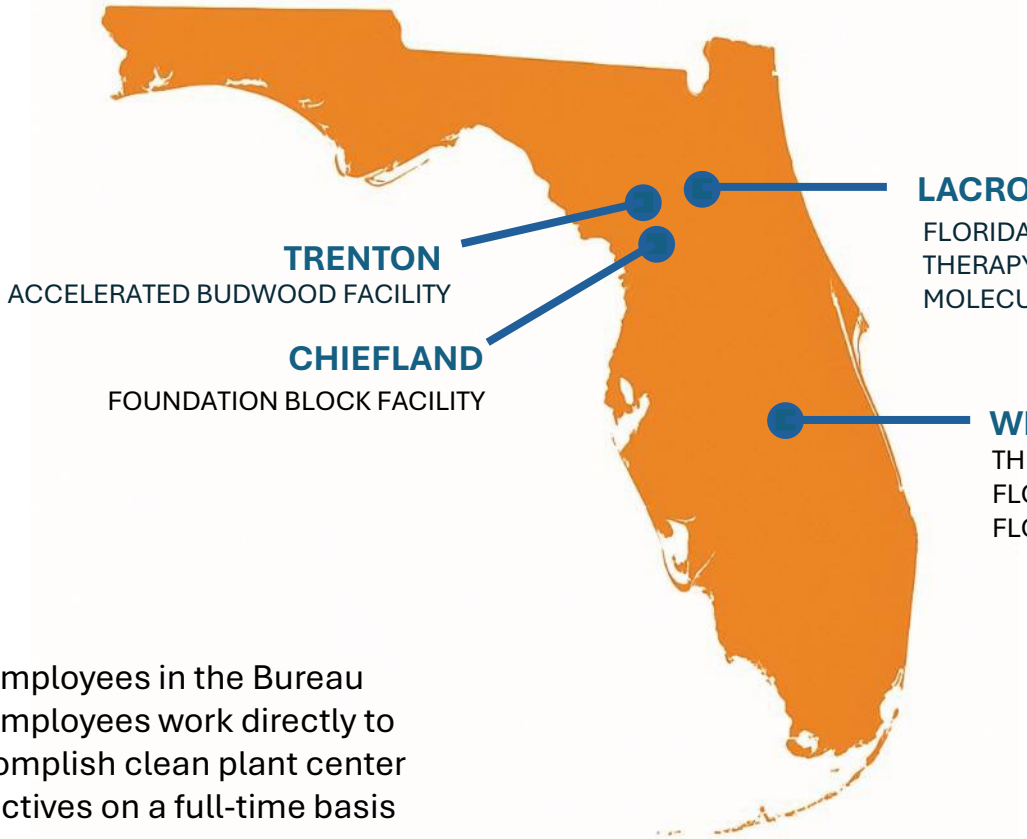
TC: Tissue culture
STG: Shoot-tip grafting



NCPN – Citrus Florida

Maria Brenelli – Biological Administrator III
Justin Ezell – Bureau Chief

Bureau of Citrus Budwood Registration Budwood Facilities around the state



LACROSSE
FLORIDA CITRUS REPOSITORY QUARANTINE FACILITY
THERAPY FOR OUT OF STATE VARIETIES
MOLECULAR DIAGNOSTICS LABS

TRENTON
ACCELERATED BUDWOOD FACILITY

CHIEFLAND
FOUNDATION BLOCK FACILITY

WINTER HAVEN
THERAPY FOR IN STATE VARIETIES
FLORIDA CITRUS NURSERY INSPECTION
FLORIDA CITRUS ARBORETUM

- 45 Employees in the Bureau
- 20 Employees work directly to accomplish clean plant center objectives on a full-time basis

DIVISION of PLANT INDUSTRY

Florida Citrus Foundation – Chiefland, FL

Aerial View of Greenhouse Facility

Facility Highlights:

- Built in 2006, expanded twice to support more varieties.
- 5 greenhouses >> 3.5 acres.

Tree Collection:

- 2,235 trees from 1,198 distinct varieties.
- 2,500 total tree spots.
- 414,285 bud eyes distributed since July 2022.
- All trees tested annually.

Backup Foundation House – LaCrosse, FL

Funds allocation 2023-24: Foundation Greenhouse Re-Roof Project

$500,000 Project

- NCPN funded 9% of cost
- State funds 91%

Impact: Increased light levels have significantly improved trees growth.

Florida Citrus Foundation - Seed Source

- Established in 2019
- Certified seed source trees – budwood from Chiefland Foundation
- 654 trees, 61 varieties
- Possibly supply seed to industry in the future

Program priorities & Strategic Considerations

Foundation Selectivity:

- Narrowing the number of varieties in care to keep cost down.

Emerging Material Types:

- Addressing protocols and transgenic citrus from inside and outside the state.

Collaboration Across Centers:

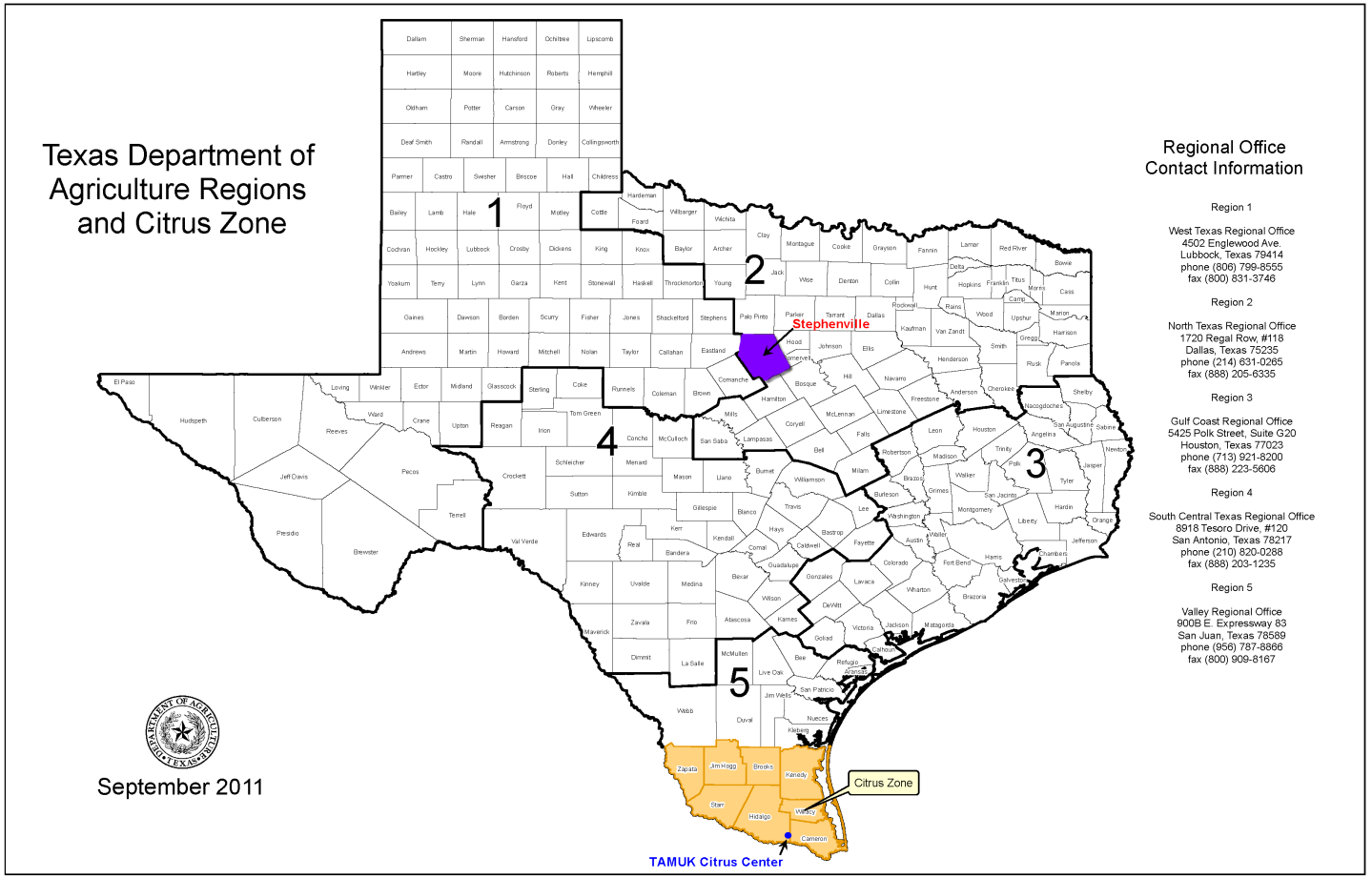
- Coordinating to avoid duplicated therapy cost and ensure NCPN isn't funding the same variety multiple times.

7721 Project: *"Enhancing Citrus Germplasm Mobility: Bridging California and Florida with Advanced qPCR Testing for Exotic and Endemic Pathogens to Accelerate Research Projects and Meet Industry Needs"* (Led by University of California, Riverside)

DIVISION of PLANT INDUSTRY

NCPN-Citrus Texas

Dr. Mamoudou Sétamou, Executive Director
Mark Vanness, Program Coordinator



TAMUK Citrus Center – Weslaco, Texas

Foundation screenhouses

Stephenville, TX Greenhouse

Expanded collection of key citrus varieties for Texas

Facilities/Improvements

- New heating systems were added in the screenhouses for additional protection in the event of another, more severe killing freeze in FY ‘25.

- Foundation Screenhouse that was destroyed in May, 2021 was rebuilt with NCPN funding, allowing for additional Foundation tree space that was lost in FY ‘25.

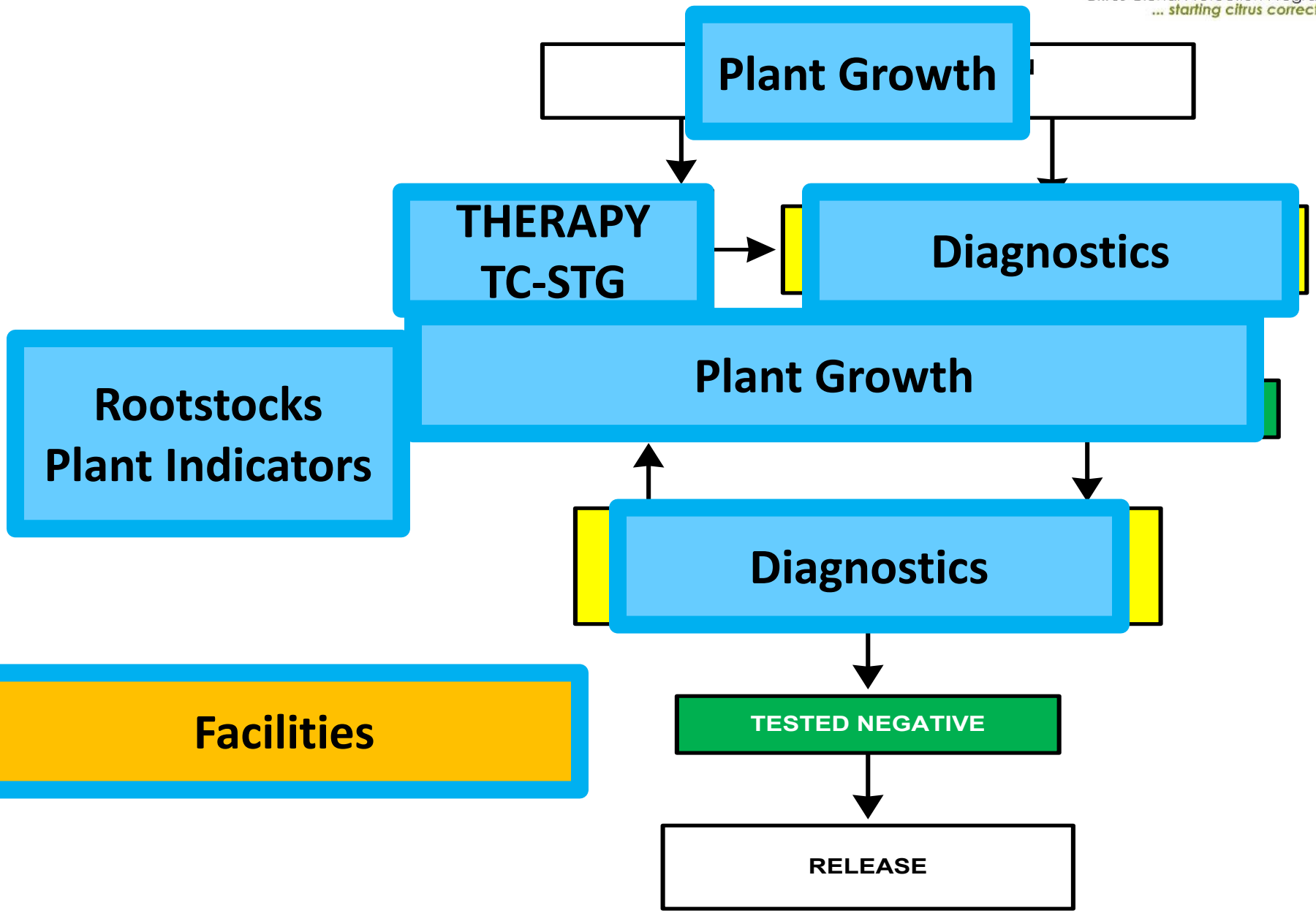
Budwood Customer List

Alabama-Georgia-Louisiana

Company	Location	Contact
Floradel Citrus Nursery	Foley, AL	Chase Cooper
Bee Nursery	Ray City, GA	Hershell Boyd
Ben & Ben Becnel Nursery	Belle Chasse, LA	Ben Becnel
Becnel Citrus Company	Belle Chasse, LA	Patrick Becnel
Star Nursery	Belle Chasse, LA	Dawn Camardelle
Sam Stokes Nursery LLC	Forest Hill, LA	Michael Lyles

TC: Tissue culture
STG: Shoot-tip grafting

Citrus Clonal Protection Program
... starting citrus correctly



CCPP Tissue Culture Projects - *In Vitro* Plant Propagation

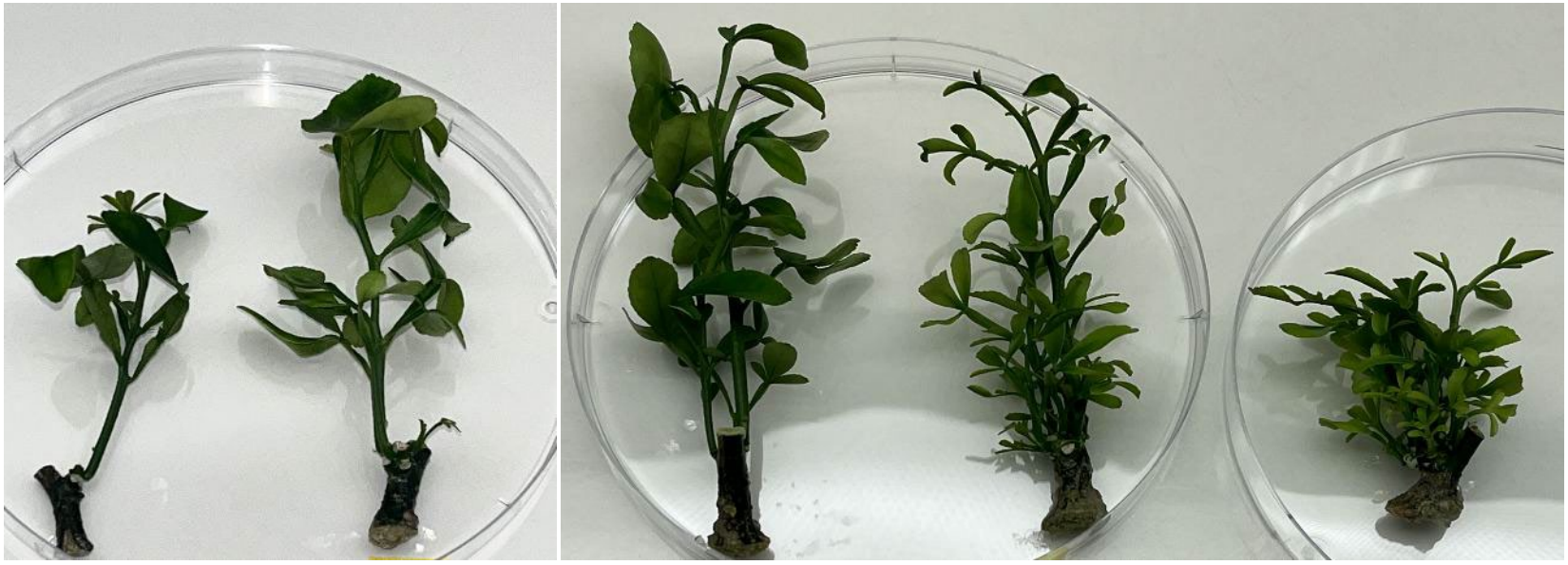
Dr. Paulina Quijia-Lamina

MINERAL COMPOSITION
MS (Murashige and Skoog Salts)
DKW (Driver and Kuniyuki Walnut Medium)

OTHER COMPONENTS	VITAMINS/AMINOACIDS
Sucrose	Thiamine
Ascorbic acid	Myo-inositol
Citric acid	Nicotinic acid
Agar	Pyridoxine-HCl
	Glycine

PLANT GROWTH REGULATORS
Auxins-NAA
Cytokines

CYTOKINES	
6-Benzylaminopurine	BA
Kinetin	Kin
6-(γ,γ-Dimethylallylamino) purine	2-iP
Meta-Topolin	mT



Pre-transplant Rooting

Dr. Paulina Quijia-Lamina

Citrus Clonal Protection Program
... starting citrus correctly

Rough Lemon **Citron**

In vitro rooting percentage of rough lemon and citron shoots under different rooting treatments after 4 weeks of culture

Citron **Rough Lemon**

Acclimatization to natural environment

CCPP-Tissue Culture Projects

Seeds are amazing evolutionary tools however, for citrus clean plant center they are a pain….$$ & Care

1. Field plots (time, land, care, disease test)
2. Fruit once per year
3. Seed maturation varies per species
4. Seed extraction, treatment, storage
5. Viability & germination rate fluctuates with time and species
6. No parameters known for citrus seed storage
7. Some new rootstock hybrids produce low or no seed

Seed Type	Drying Tolerance	Storage Potential	Examples
Orthodox	✅ Can survive desiccation	✅ Long-term storage	Wheat, Rice, Maize, Beans
Recalcitrant	❌ Cannot dry out	❌ Short-lived, must stay moist	Mango, Avocado, Cocoa
Non-Orthodox	⚖️ Partially tolerant	⚖️ Intermediate storage	Citrus, Coffee, Oil Palm

Chat-GPT

Citrus Seed Drying, Storage & Germination

Dr. Paulina Quijia-Lamina

- **Moisture content:** The oven method
  - Fresh weight
  - Dry weight: 105 C for 16-18 h.
  - Moisture content (%)=
    Fresh Weight-Dry Weight\Dry Weight X 100
- **Drying: Air drying**
  - Temperature : 19.11 °C
  - RH: 43.68%
- ***In vitro* seed germination** CCPP protocol
  - 20% Bleach for 35 min
  - Dark at 28 °C

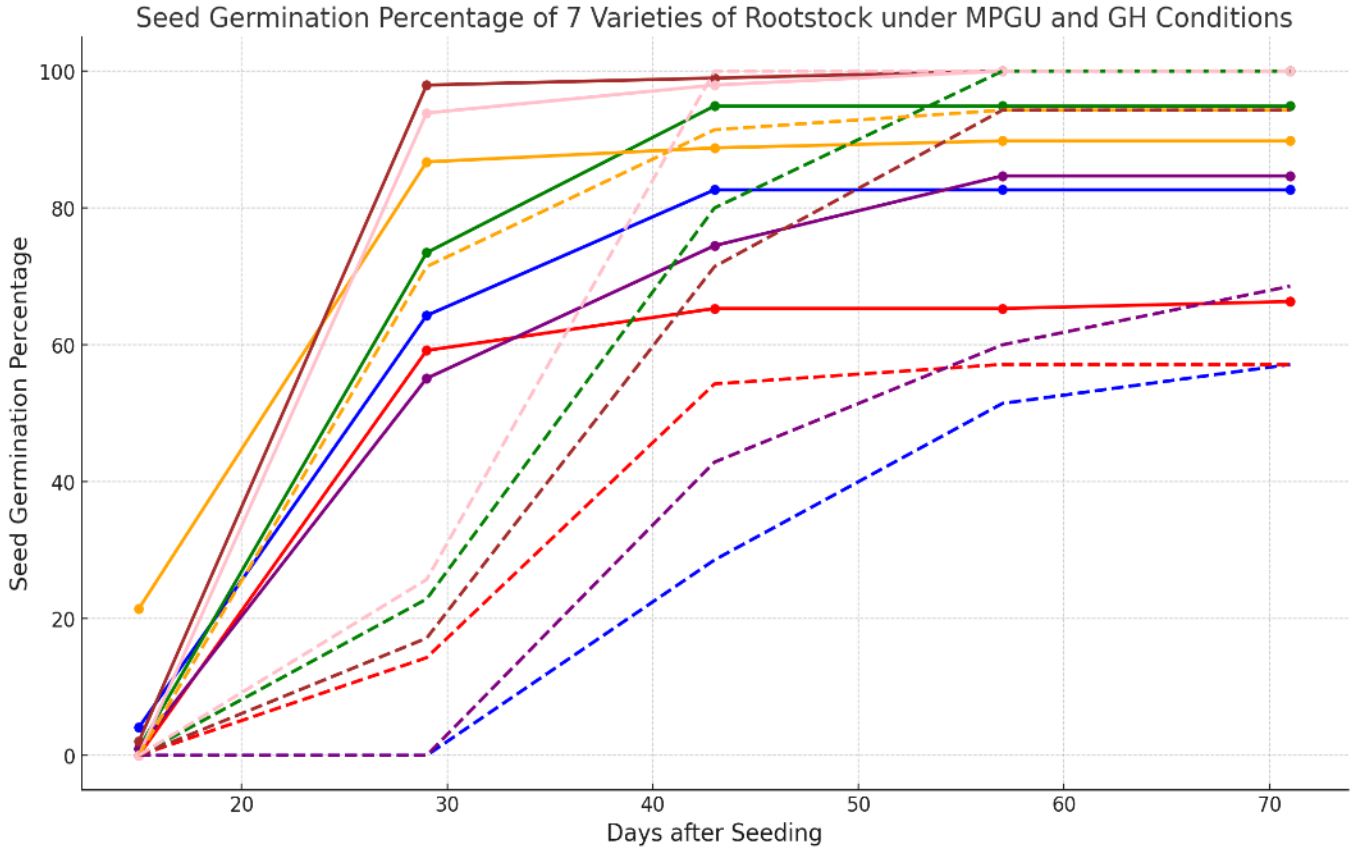
T °C	MOISTURE CONTENT			
8 °C	40 %	30 %	20%	10%
2 °C	40 %	30 %	20%	10%

Citrus Clonal Protection Program
... starting citrus correctly

Dr. Deborah Pagliaccia

MPGU – CEA - CCPP

MPGU: Rapid and high germination rates by day 43.
GH: Gradual germination high rates by day 71.

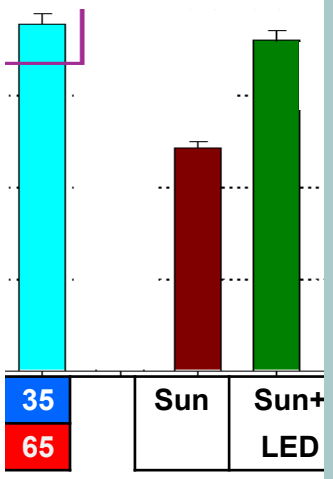


Light quality and quantity are critical for plant growth, influencing growth rates, photomorphogenesis, and tissue composition.

GR	W Only	W + BR	BGR	W + BR	BGR	BR
PH 1	PH 2	PH 3	PH 4	PH 5	PH 6	PH 7
0 %	7-8	7-8	15	35	35	35
35 %	35	20	16	14	30	1
50-55 %	50-55	65-70	65-70	40-45	35-40	65-70

MPGU: 14DLI

GH



MPGU

%: 35

%: 65

GH

Sun+

LED

GH

Sun

Light wavelength and temperature manipulation affect disease symptom expression in citrus indicator plants

Stacey Comstock
PhD student

Rack 1

95R:5B
85R:15B

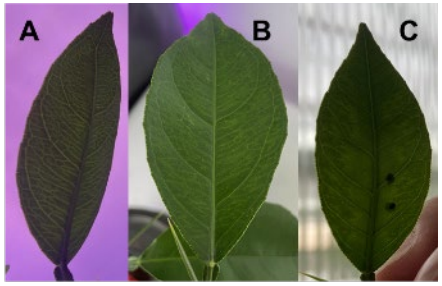
Rack 2

75R:25B

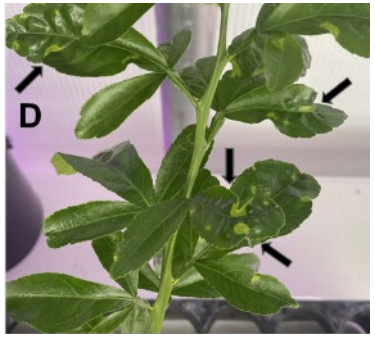
65R:35B

Rack 3

Full Spectrum



CiVA: A, B "young leaf pattern", C "oak leaf pattern"



CTLV: D, chlorotic spots

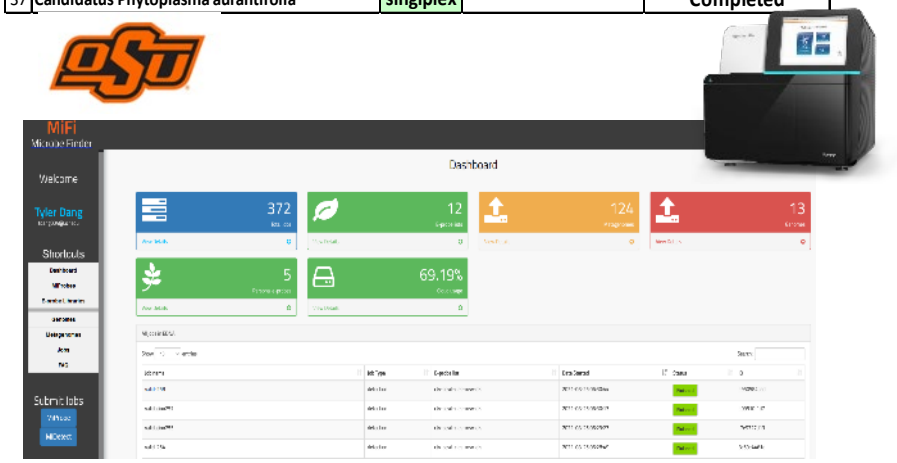
Standard Greenhouse

Real-time multiplex-in vitro diagnostic assays-48 targets (more in development)

Dr. Fatima Osman
Dr. Sohrab Bodaghi

Viruses		qPCR Test		HTS Test
1	Citrus tristeza virus (CTV)	singlplex	Mulitplex (CTV, CPsV, CLBV)	Completed & In use
2	Citrus psorosis virus(CPsV)	singlplex		Completed
3	Citrus leaf blotch virus (CLBV) syn. Dweet mottle virus	singlplex		Completed
4	Citrus tatter leaf virus (syn. Apple stem grooving virus) (CTLV)	singlplex	Mulitplex (CTLV, CVEVV,CYVV)	Completed
5	Citrus vein enation virus (CVEV)	singlplex		Completed
6	Citrus yellow Vein virus (CYVV)	singlplex		
7	Citrus sudden death-associated virus (CSDaV)	singlplex	Mulitplex (SDaV, CJLV,CVLV)	
8	Citrus jingmen-like virus	singlplex		
9	Citrus virga-like virus (CVLV)	singlplex		
10	Citrus chlorotic dwarf-associated virus (CCDaV)	singlplex	Multiplex DNA assay (CCDaV, CYMV)	
11	Citrus yellow mosaic virus(CYMV)	singlplex		
12	Satsuma dwarf virus (SDV)	singlplex	Mulitplex (SDV,CiMV, NIMV)	
13	Citrus mosaic badnavirus (CiMV)	singlplex		
14	Natsudaidai dwarf virus (NDV)	singlplex		
15	Navel infectious mottling virus (NIMV)	singlplex		
16	Indian citrus ring-spot virus (ICRSV)	singlplex		
17	Hyuganatsu virus	singlplex		
18	Citrus yellow vein clearing virus-Mandarivirus (CYVCV.2)	singlplex		
19	Citrus leprosis cilevirus C (CiLV-C)	singlplex		
20	Citrus leprosis dichorhavirus N (CiLV-N)	singlplex		
21	Citrus variegation illarvirus (CVV) (syn. Citrus crinkly leaf virus)	singlplex		Completed
22	Citrus leaf rugose ilarvirus virus(CLRV)/	singlplex		
23	Citrus chlorotic dwarf-associated virus (CCDaV)	singlplex		
24	Citrus virus A	singlplex		Completed
25	Citrus yellow mottle associated-Mandarivirus (CYMoV)	singlplex		
26	Citrus Concave gum associated virus	singlplex		Completed

Viroids		qPCR Test		HTS Test
27	Citrus exocortis Non-Apsca viroid	singlplex	Multiplex Non-Apsca vioroids (CEVd, HSVd, CBCVd)	Completed & In use
28	Hop stunt Non-Apsca viroid (HSVd), citrus variants of HSVd, CVd-IIa, -IIb & -IIc	singlplex		Completed
29	Citrus bark cracking Non-Apsca viroid (CBCVd)	singlplex		
30	Citrus bent leaf Apsca viroid (CBLVd)	singlplex		
31	Citrus Apsca viroid VI (-OS)	singlplex		
32	Citrus dwarfing Apsca viroid (CDVd)	singlplex		
33	Citrus Apsca viroid V (CVdV)	singlplex		
34	Citrus Apsca viroid VII (CVdVII)	singlplex		
Prokaryotes				
35	Candidatus Liberibacter sp.	singlplex		Completed
36	Spiroplasma citri	singlplex		Completed
37	Candidatus Phytoplasma aurantifolia	singlplex		Completed



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Chambers et al. 2022. J. Virological Methods 299: 114330
Osman et al. 2015. J. Virological Methods 220: 64-75.
Osman et al. 2017. J. Virological Methods 245: 40-52.
Dang et al. 2022. Methods Molecular Biology, Viroids. Vol. 2316. Springer.
Dang et al. 2023. PhytoFrontiers™3:1, 113-123
Nascimento et al. 2025 PhytoFrontiers™

| 2025 • 5:243-253 https://doi.org/10.1094/PHYTOFR-12-24-0140-FI

PhytoFrontiers™
An Open Access Journal from The American Phytopathological Society

Research

Development and Validation of a Suite of E-Probes for Electronic Diagnostic Nucleic Acid Analysis (EDNA) for 20 Graft-Transmissible Pathogens of Citrus Using MiFi and Blind Ring Testing Among Novice Users

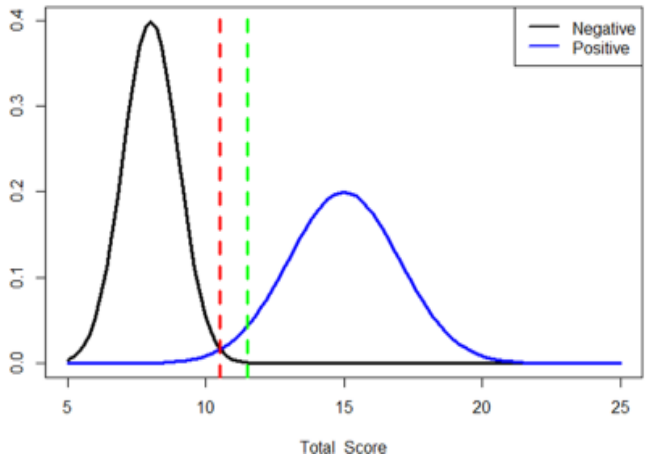
D. M. Nascimento[1,2] | S. Bodaghi[3] | H. Wang[4] | M. R. Ribeiro-Junior[1,2] | R. Campos[3] | T. Dang[3] | F. Osman[5] | J. Habiger[4] | A. S. Espindola[1,2] | G. Vidalakis[3,†] | K. F. Cardwell[1,2,†]

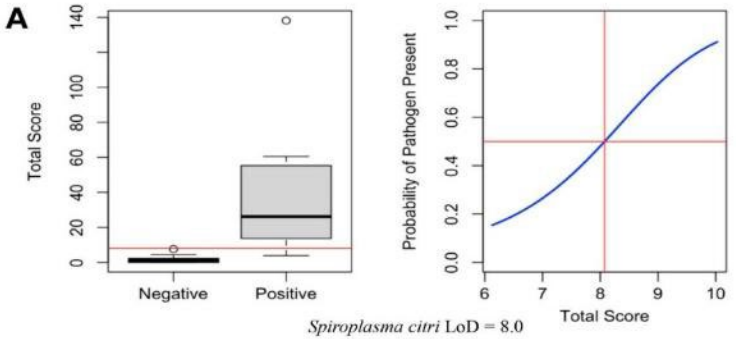
E-probe validation – proposed equations for LOD in HTS data

Formula of LODs: Statistical LOD is defined by a Bayesian decision boundary when we use the quadratic discriminate analysis (QDA) to classify known positive and negative sample Scores

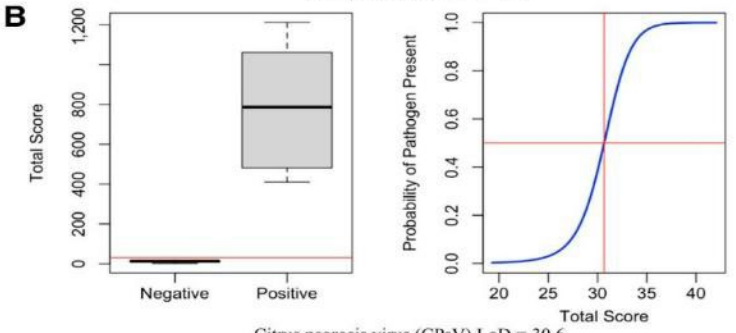
$$LOD_{50/50} = \frac{\left(\frac{\mu_2}{\sigma_2^2} - \frac{\mu_1}{\sigma_1^2}\right) - \sqrt{\frac{(\mu_1-\mu_2)^2}{\sigma_1^2\sigma_2^2} - \left(\frac{1}{\sigma_2^2} - \frac{1}{\sigma_1^2}\right) \times 2log\frac{\sigma_2^2}{\sigma_1^2}}}{\left(\frac{1}{\sigma_2^2} - \frac{1}{\sigma_1^2}\right)} \qquad LOD_{80/20} = \frac{\left(\frac{\mu_2}{\sigma_2^2} - \frac{\mu_1}{\sigma_1^2}\right) - \sqrt{\frac{(\mu_1-\mu_2)^2}{\sigma_1^2\sigma_2^2} - \left(\frac{1}{\sigma_2^2} - \frac{1}{\sigma_1^2}\right) \times 2log\frac{4\sigma_2^2}{\sigma_1^2}}}{\left(\frac{1}{\sigma_2^2} - \frac{1}{\sigma_1^2}\right)}$$

Where μ_1 and σ_1^2 are mean and variance of Total Score for the negative group. Similarly, μ_2 and σ_2^2 are mean and variance of Total Score for the positive group.

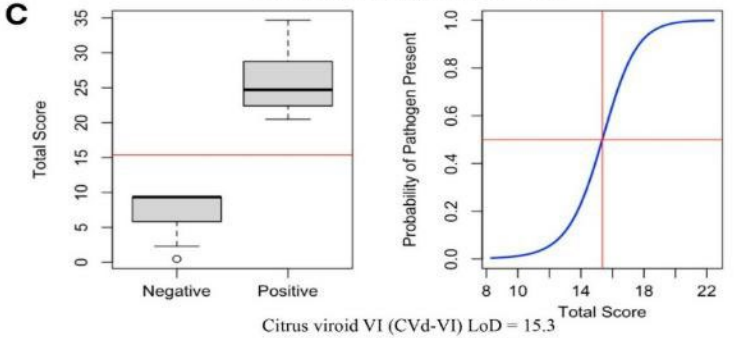




Spiroplasma citri LoD = 8.0



Citrus psorosis virus (CPsV) LoD = 30.6



Citrus viroid VI (CVd-VI) LoD = 15.3

Table 3. CCPP VI index: Diseases, pathogens, and testing procedure

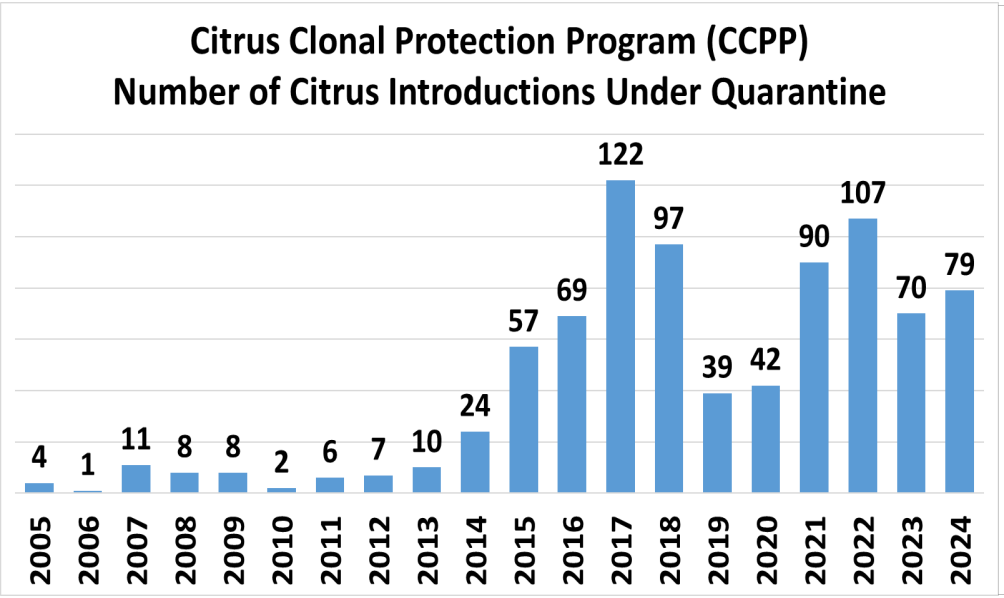
DISEASE[1]	PATHOGEN	BIOLOGICAL INDEXING & LABORATORY TESTING
	Viruses	
Tristeza, Quick Decline, Stem pitting, & Seedling yellows	1. Citrus tristeza virus (CTV)	Mexican lime[2], RT-qPCR[3, 4], HTS-EDNA[12]
Psorosis A & B (syn. ring spot)	2. Citrus psorosis virus (CPsV)	Sweet orange, Dweet tangor, RT-qPCR, HTS-EDNA
Leaf blotch Dweet mottle	3. Citrus leaf blotch virus (CLBV) (syn. Dweet mottle virus) 4. Citrus leaf blotch virus 2 (CLBV-2)	Dweet tangor, RT-qPCR, HTS-EDNA (CLBV)
Infectious variegation (syn. crinkly leaf) Leaf rugose	5. Citrus variegation virus (CVV) (citrus crinkly leaf virus) 6. Citrus leaf rugose virus (CLRV)	Sour orange, 'Etrog' citron[5]/RL[6], RT-qPCR, HTS-EDNA (CVV)
Tatter leaf Citrange stunt Pummelo leaf yellow mottle mosaic	7. Apple stem grooving virus (ASGV) (syn. citrus tatter leaf virus)	Rusk citrange/RL, *Citrus excelsa*, RT-qPCR, HTS-EDNA
Vein enation	8. Citrus vein enation virus (CVEV)	Mexican lime, Sour orange, RT-qPCR, HTS-EDNA

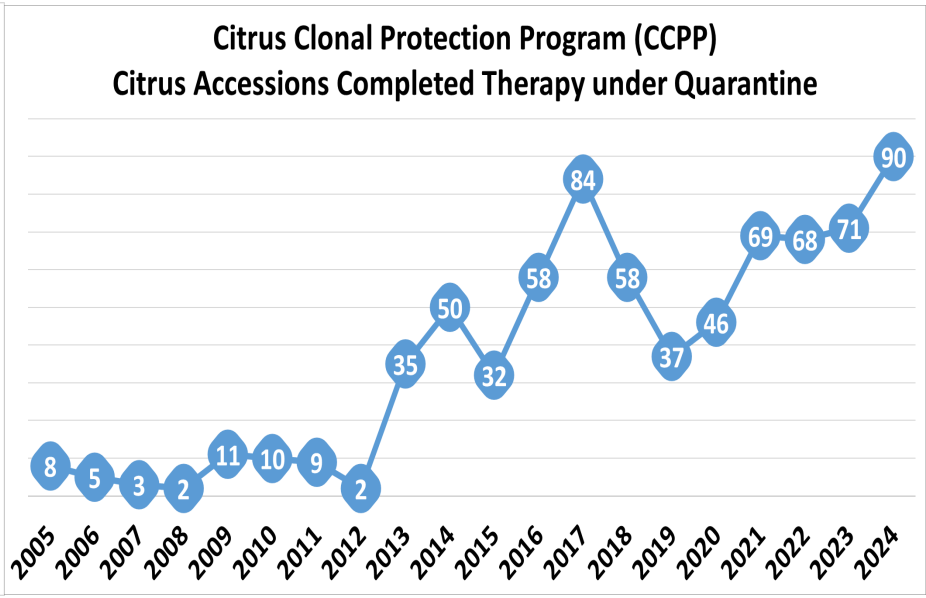
CFR § 319.19. Citrus and its relatives are prohibited from entering the USA

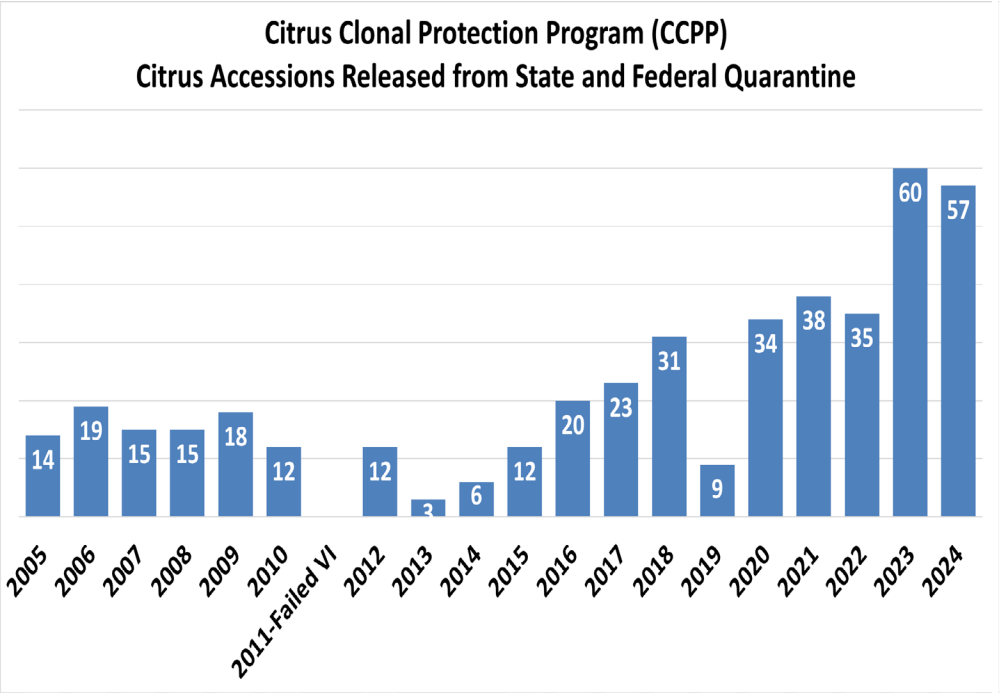
"…in order to prevent the introduction into the United States of ...citrus diseases…"

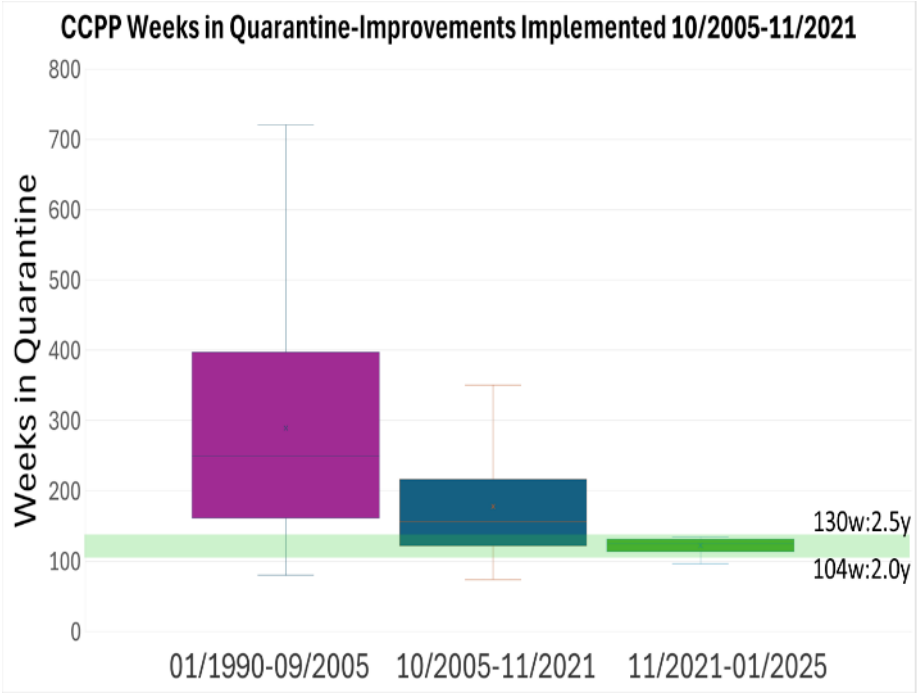
*"…may be imported into the United States …under the conditions specified in a controlled import permit issued in accordance with **CFR § 319.6…**"*

CCPP-Quarantine Capacity









Upcoming Varieties

CCPP Citrus Varieties*

Introduction Option	Varieties Under Quarantine	Quarantine Released Varieties	Total
Proprietary	37	138	175
Public Domain WITH Financial Support from Requester	4	8	12
Public Domain WITHOUT Financial Support from Requester	43	210	253
Research-HLB Resistant Varieties-CRB & USDA MAC Funded	0	68	68
Specific Agreement	2	30	32
Variety Diversity	0	10	10
UC Riverside-Citrus Variety Collection	5	10	15
UCR-CBP-UC Riverside-Citrus Breeding Program-NOT FOR RELEASE	9	58	67
National Clean Plant Network (NCPN)	9	4	13
Research-Variety Germplasm	1	13	14
USDA-ARS-Citrus Breeding Program-USDA RELEASED	5	1	6
USDA-ARS-Research Materials	17	8	25
Fruitmentor-CRB Project	3	8	11
UF-CREC-Citrus Breeding Program-Under Material Transfer Agreement (MTA)-NOT FOR RELEASE	44	2	46
USDA-Citrus Breeding Program-Under Specific Agreement - USDA RELEASE PENDING	24	2	26
CCPP-ReIndex	30	36	66
USDA-ARS-National Clonal Germplasm Repository for Citrus and Dates (NCGRCD)	13	10	23
UC Riverside-Research Materials	0	7	7
UCR-CBP-UC Riverside-Citrus Breeding Program-RELEASED	0	0	0
	246	623	869

* Number of varieties since 04/2015.

National Citrus Nursery Association NCNA

Home | Leadership & Associations | Resources | News & Events | Contact | Members

Dr. Deborah Pagliaccia

UC RIVERSIDE UNIVERSITY OF CALIFORNIA

PPA 7721 project titled “Enhancing U.S. Citrus Nursery Sustainability: Collaborative Stakeholder Alignment & Engagement”

Welcome to the National Citrus Nursery Association (NCNA)

Promoting Best Practices for Citrus Nursery

Enhancing U.S. Citrus Nursery Sustainability

A USDA APHIS-Funded Initiative

This initiative, funded by USDA APHIS, is dedicated to strengthening the U.S. citrus nursery industry through stakeholder collaboration, sustainable innovation, and best practices adoption. Learn how we're supporting clean, disease-free citrus production nationwide.

Learn More

USDA APHIS

1. **Regulatory Reform & Compliance**

“one set of rules please”

2. Nursery Certification Program Modernization

3. Pest Prevention & Risk Management

4. E-Commerce & Unregulated Citrus Plant Material

5. Research & Education Needs

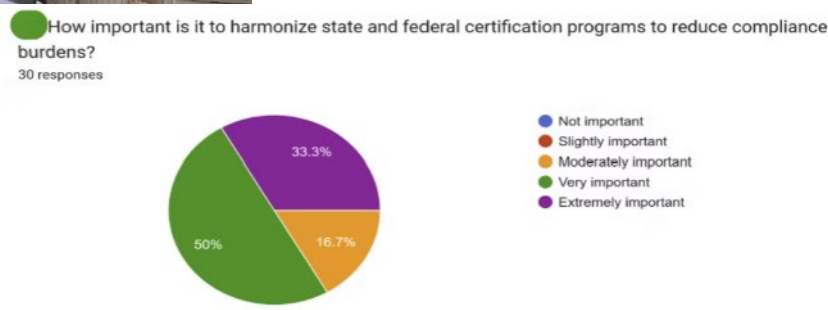
6. Federal & State Coordination

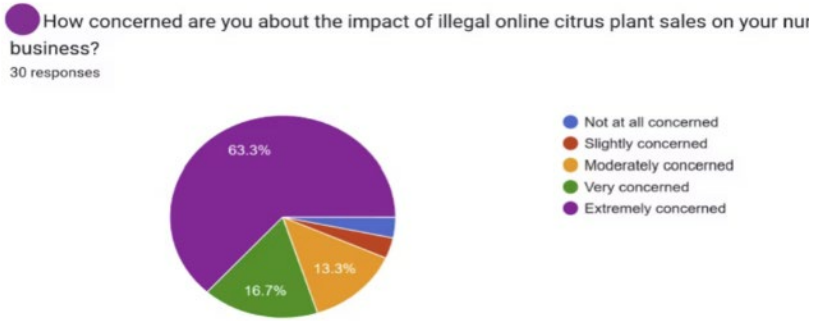
7. Industry Sustainability & Innovation

United States Department of Agriculture
Animal and Plant Health Inspection Service
Plant Protection and Quarantine

INTERSTATE MOVEMENT OF CITRUS NURSERY STOCK FROM AREAS QUARANTINED FOR CITRUS CANKER, CITRUS GREENING, AND/OR ASIAN CITRUS PSYLLID

November 25, 2019





A Systems Approach

Pathogen Tested Budwood

Indoor Citrus Nursery Production

Citrus Nursery Stock Pest Cleanliness Program

Healthy Orchard Programs

Citrus Clonal Protection Program
... starting citrus correctly

University of California
Agriculture and Natural Resources

Citrus Pest Detection Program
Central California Tristeza Eradication Agency

CITRUS PEST & DISEASE PREVENTION PROGRAM

Thank you

vidalg@ucr.edu
www.labtofarm.org

Three years in: Building a fully functional Arkansas Center

U of A DIVISION OF AGRICULTURE
RESEARCH & EXTENSION
UNIVERSITY OF ARKANSAS SYSTEM

THE ARKANSAS
CLEAN PLANT CENTER

The basics

- ~ 1600sqft of newly renovated lab space + ~ 400sqft for equipment
- 800K from USDA (no NCPN funds used) for equipment
- ~1.6 M by Arkansas Experimental Station; lab and greenhouse renovations, construction of the foundation greenhouse (closed system)
- Fully functional vertical Center as of November 2022

THE ARKANSAS
CLEAN PLANT CENTER

Receiving lab

Sample preparation

1) Tissue inspection, labelling, storage and sampling
2) Initial extraction (sample homogenization)

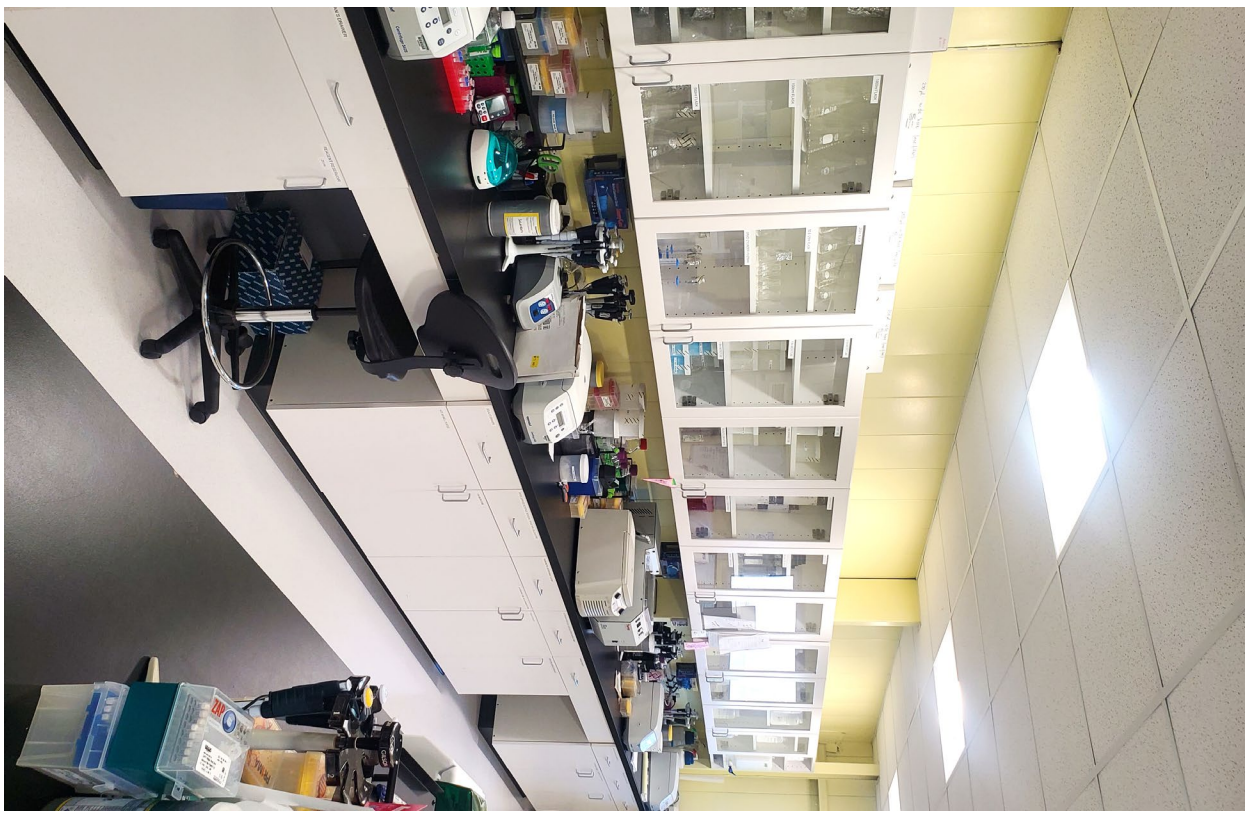
TC media preparation

Molecular biology lab

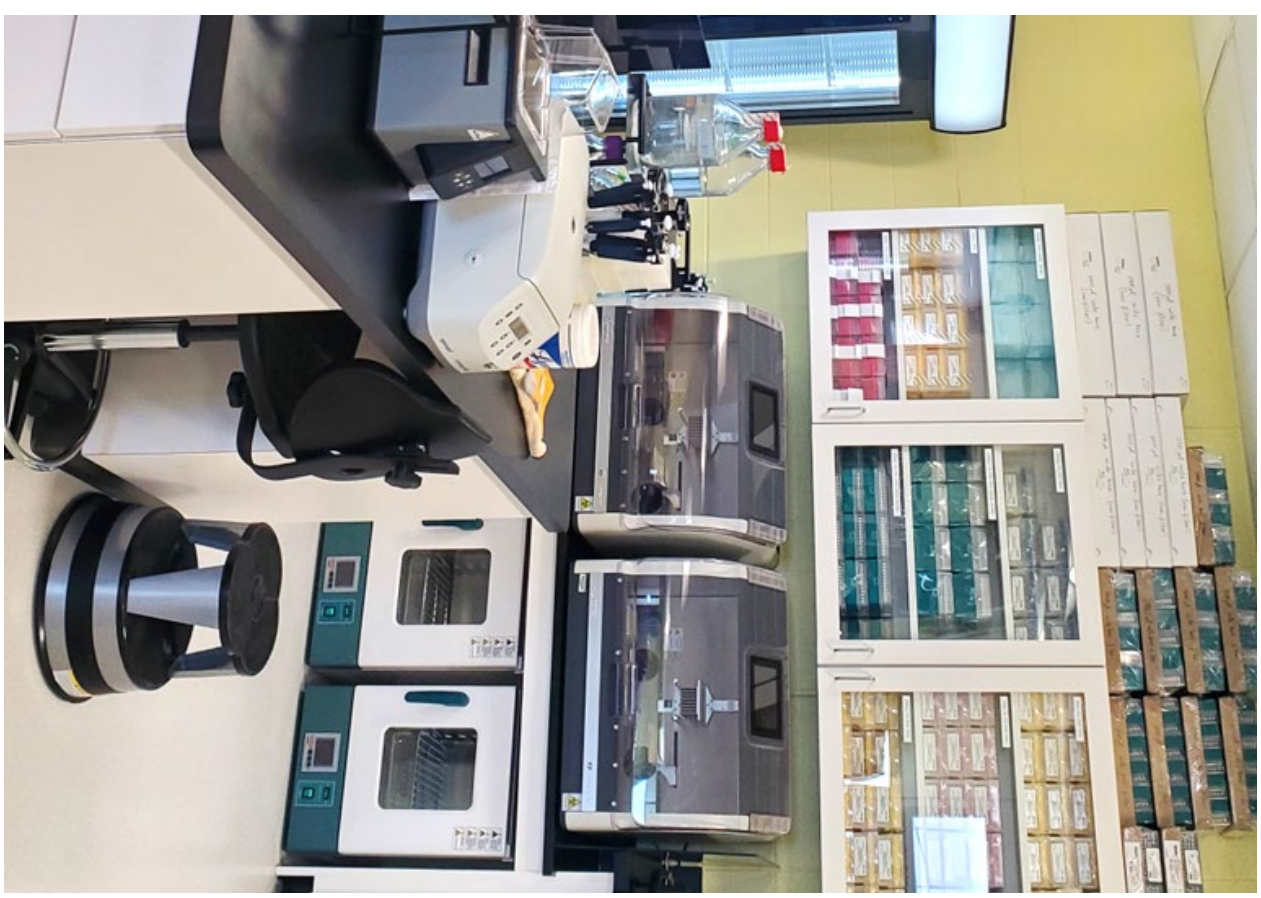
1) TNA/RNA purification – manual and automated

2) PCR mastermix preparation – manual pipetting

Dedicated work benches for each personnel

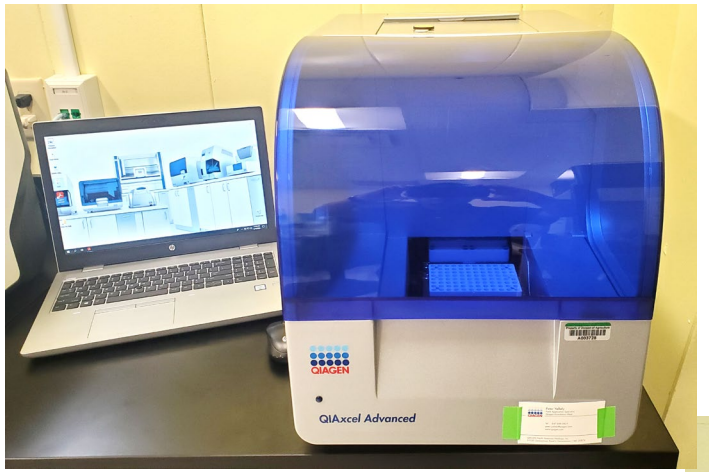


Fully automated TNA extraction



PCR lab

10 PCR machines - 8 endpoint (3 not in photo), 2 quantitative



Capillary electrophoresis

Dedicated PCR workstation

Pipetting robotic system

Freezer and stock room

1) Conventional gel electrophoresis and documentation (back-up)

HTS room

Dedicated for library prep and HTS processing only.

Brand new Next Seq ™

TC lab

1) Virus elimination, shoot-tip culture and transfers.
2) One-way street to the autoclaved media.
3) Microscopy and imaging of the grown seedlings or plantlets.

Growth chambers

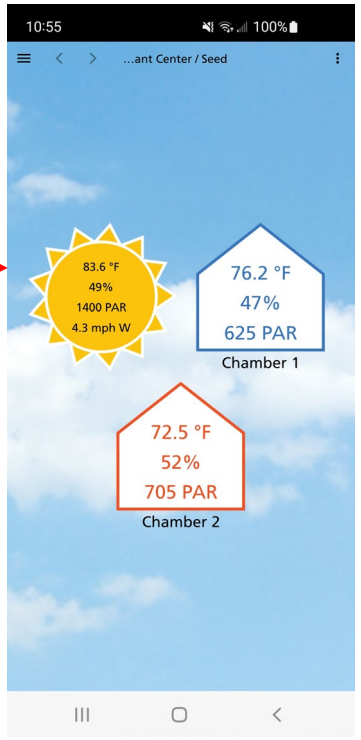
1) Heat treatment of the introductions.
2) Maintenance of meristem-derived daughter plants from introductions, post virus clean-up.
3) Dedicated growth chamber for CIP and in-vitro maintenance of plantlets.

G1 Greenhouse

Quarantine style greenhouse - ~600sqf, fully A/C

State-of-the-art green house – minimal to no human interventions

Smart phone control



Fully automated fertigation system

Seed software allows remote control from anywhere.

Why are we here?

Mission

Safeguard and improve fruit crop productivity and sustainability in the United States by providing clean plants.

Vision

Enhance the productivity of United States fruit crops through the provision of clean plants. We aim to foster innovation, bolster the growth of the fruit industry, and promote sustainable agricultural practices.

THE ARKANSAS
CLEAN PLANT CENTER

Core values

- **Quality**: Ensure highest standards in providing clean, disease-free plant material.
- **Innovation**: Seek and apply innovative solutions and technologies to advance fruit crop health.
- **Collaboration**: Build strong partnerships with other Clean Plant Centers, producers and industry stakeholders by sharing knowledge and resources to achieve shared objectives.
- **Education and Outreach**: Inform stakeholders on the benefits of using clean plants.
- **Adaptability**: Be flexible and responsive to evolving challenges and opportunities in specialty crops.
- **Integrity and Transparency**: Conduct operations with integrity, openness, and accountability to foster trust amongst stakeholders and the National Clean Plant Network.

THE ARKANSAS
CLEAN PLANT CENTER

The people

Dr. Dan Edward Veloso Villamor
Center Operations
Diagnostics & Bioinformatics

Dr. Shivani Singh
Therapy, Tissue Culture

Dr. Bolaji Osundahunsi
Diagnostics & Bioinformatics

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CLEAN PLANT CENTER

Logistics

- Started clean up process in January 2023: strawberry, blackberry, raspberry and elderberry
  - 22 accessions in clean-ups as of August 2025: one strawberry and two elderberry selections are projected to be released to the breeder/sponsor by the end of this year (total of four rounds of HTS)
- Serving industry (including State Depts of Ag – 15 clients on diagnostics for certification and exports

  - In 2022: ~7700 PCRs, 3000 ELISAs, 24 HTS

    2023: ~7800 PCRs, 1400 ELISAs, 44 HTS

    2024: ~10000 PCRs, 78 HTS

    2025: target ~10000 PCRs, 150 HTS
- Obtained CIP in August 2025

THE ARKANSAS
CLEAN PLANT CENTER

360 assessment – 2022

Top attributes:

- Diversity
- Effective team
- Ethics and values
- Integrity and trust
- Organization
- Perseverance
- Priority setting
- Problem solving
- Strategic agility

THE ARKANSAS
CLEAN PLANT CENTER

Off the beaten path….
Phantom pathogen review & EPPO regulatory update

- Global collaboration: 185+ scientists, 40+ countries, 120 phantom agents across 10 NCPN genera

- Raspberry leaf curl and Strawberry latent C under review for removal from EPPO A1 list → grafting restrictions lifted - EPPO Reporting Service No. 05–2025 (article 2025/130)

- EPPO panel on Phytosanitary Measures to implement deactivations and update certification guidelines

- **Significance:** Streamlines certification, reduces grafting burdens, accelerates exchange of virus-free germplasm

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CLEAN PLANT CENTER

G1 plants, Control Import Permits and certification

- Pathogens of concern for Berries G1 differ from those required in Control Import Permits (CIPs).

- Certification programs only accept G1-level tested material as mother plants → CIP plants are not eligible.

- Working with industry to update the list of pathogens of concern; once finalized, this list will be communicated to APHIS.

- **Goals:**
  - Align testing standards across G1 and CIPs to streamline certification
  - Secure Centers from wheat head blight type incidences…

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CLEAN PLANT CENTER

ACPC CIP SOPs - four tests over two years. Why?

- **Four sampling times** were needed to detect all pathogens—no single collection was sufficient.
- **PCR failed** at times due to **primer–template mismatches**, leading to false negatives. Grafting was totally unreliable.
- **HTS outperformed** standard methods by providing **unbiased, comprehensive detection**.
- **Take-home:** HTS reduces diagnostic gaps and is a better tool for CIP than conventional detection methods

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CLEAN PLANT CENTER

Solving the problem of positive controls

- Traditional positive controls are hard to obtain, costly, and regulated

- Virus-mimicking artificial positive controls (ViMAPCs) replicate virus titer and tropism, produced in ~5 days. Available for **conventional and qPCR - viruses, fungi and nematodes**

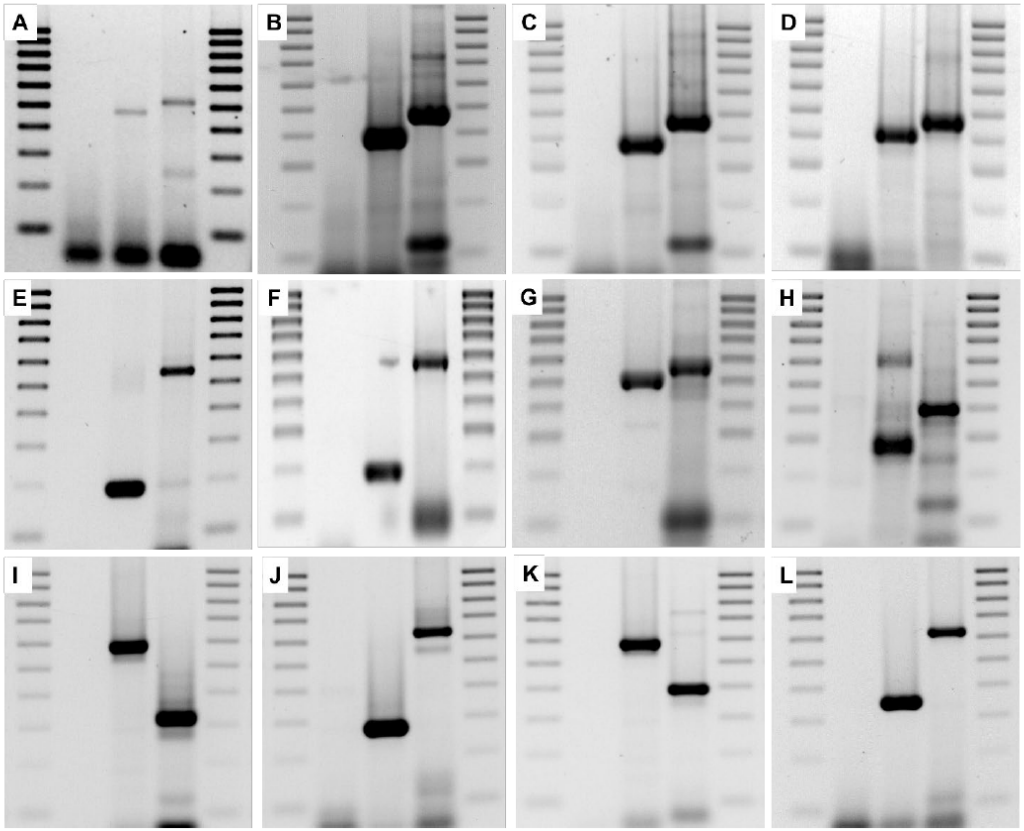
- Fast, safe, and affordable solution for reliable diagnostics and certification.

- Have developed 28 for viruses infecting berry crops and expanding

THE ARKANSAS
CLEAN PLANT CENTER

Lane 1 = 100 bp DNA marker; Lane 2 = Plant negative for target virus; Lane 3= Plant positive for target virus; Lane 4 = ViMAPC and Lane 5 = 100 bp DNA marker.

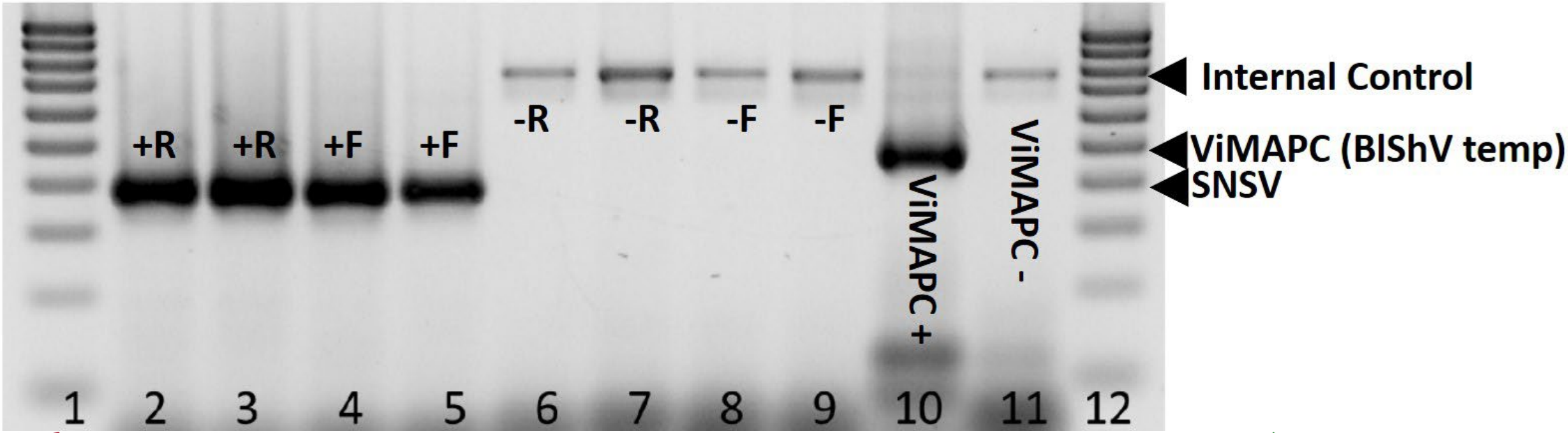
A. blackberry virus-F (BVF) using fig badnavirus-1 (FBV-1) as template; **B.** strawberry necrotic shock virus (SNSV) using blueberry shock virus (BlShV) as template; **C.** SNSV using blackberry chlorotic ringspot virus (BCRV) as template; **D.** BCRV using BlShV as template; **E.** peach rosette mosaic virus (PRMV) using tomato ringspot virus (ToRSV) as template; **F.** PRMV using tobacco ringspot virus (TRSV) as template; **G.** ToRSV using TRSV as template; **H.** rose rosette virus (RRV) using blackberry leaf mottle associated virus (BLMaV) as template; **I.** raspberry leaf blotch virus (RLBV) using RRV as template; **J.** RRV using RLBV as template; **K.** RLBV using BLMaV as template and **L.** BLMaV developed using RLBV as template.



Amplicon relative intensities:
+/- 30% of true positive

Application

Strawberry necrotic shock virus (SNSV) virus-mimicking artificial positive control (ViMAPC) developed using blueberry shock (BlShV)-infected material as template. Material infected with SNSV (*Rubus:* PI 691930 and PI 691943, *Fragaria* : PI 691784; PI 691800) and previously tested negative for the virus (*Rubus* : PI 691933 and PI 691936; *Fragaria* : PI 691758 and PI 691766) were tested alongside the ViMAPC (Lane 10) (supplementary table 3). All lanes show expected band sizes (supplementary table 1 and 2) (Thekke-Veetil et al., 2016). Lane 1 and Lane 12 represented 100 bp DNA marker.



The future of ACPC

- Strategic plan 2024-2028 completed - https://acpc.uada.edu/about/

Action items:

- Operations management
- Quality control
- LIMS
- Economic impact
- Expand HTS and bioinformatics capacity
- Strengthen virus elimination infrastructure – cryotherapy
- Enhance international collaborations and initiate discussions on global regulatory harmonization

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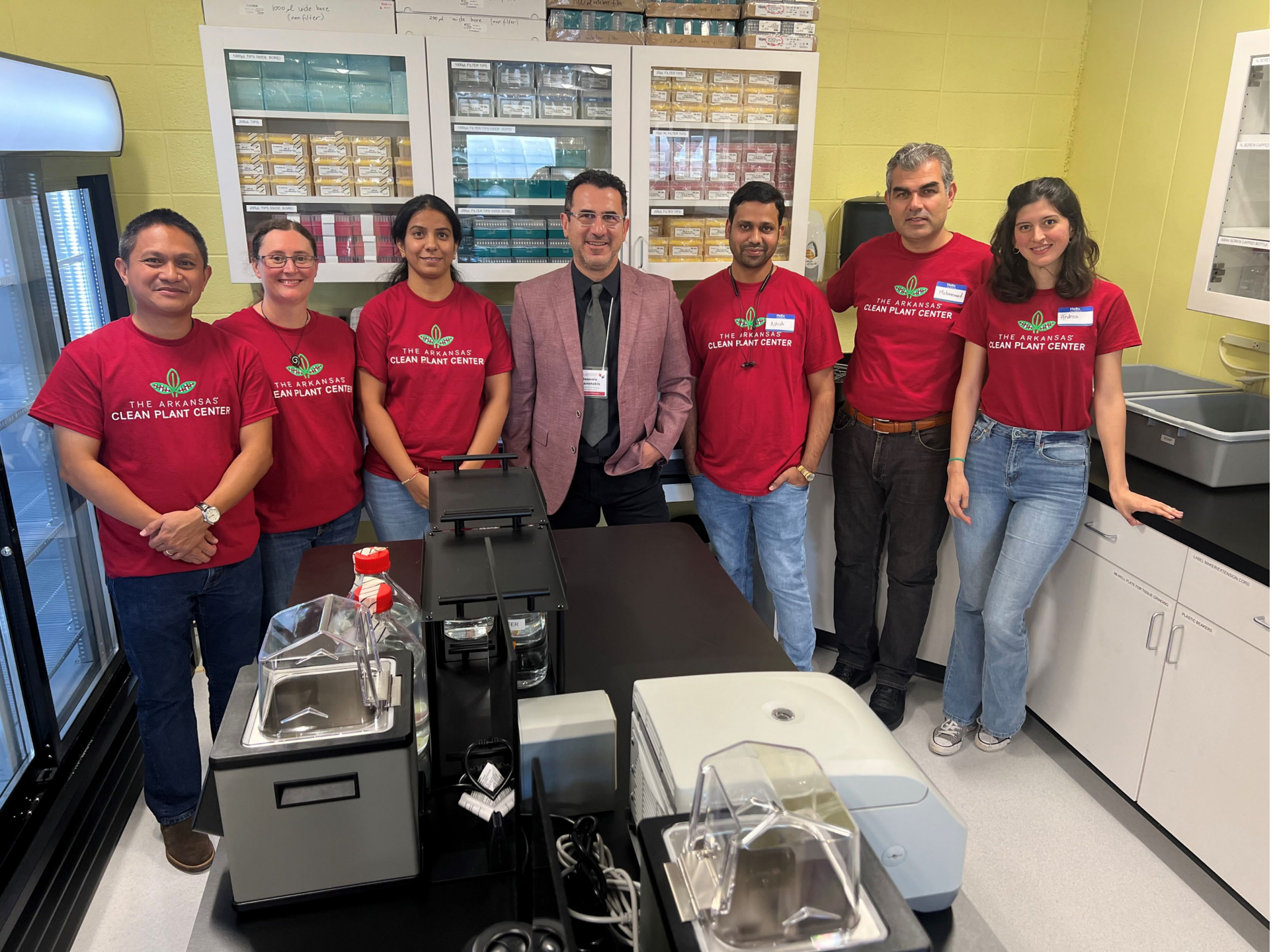
CLEAN PLANT CENTER



THE ARKANSAS
CLEAN PLANT CENTER

Update from the Oregon Clean Plant Center

(NCPN-Berries, Corvallis, OR)

Presented by: Cristian Olaya

Oregon State University, Botany and Plant Pathology Department, OSU Plant Clinic

USDA-ARS, Horticultural Crops Disease and Pest Management Research Unit, Corvallis, OR

Start clean, stay clean

NCPN National Clean Plant Network

Start clean, stay clean

Oregon State University

USDA Agricultural Research Service

NCPN-WERA-20 meeting, Raleigh/Durham, NC, September 08-10, 2025

HISTORY

- The Oregon Clean Plant Center originated at the USDA-ARS in Corvallis, Oregon, during the 1970s. Dr. Richard H. Converse began testing berry genotypes from USDA-ARS breeders Dr. Francis J. Lawrence and George Waldo ensuring virus-free stock (Willamette raspberry, Hood strawberry, and Marion blackberry).

Willamette raspberry

- In the early 1990s, Dr. Robert R. Martin expanded the clean plant program to encompass public breeders of Rubus, Fragaria, Vaccinium species to facilitate an international berry industry, supported by both federal funding and industry stakeholders. It incorporated biological indexing, serological and molecular testing, and thermotherapy. This program became a cornerstone of the international berry industry

RBDV infected raspberry. Photo R. Martin

HISTORY

- The NCPN initiative was ignited by the grape and fruit tree industries in 2005, leading to the formation of a national group to secure clean foundation cultivars. By 2008, the grape and fruit tree commodity groups were formally established under the Farm Bill (H.R. 6124, Food, Conservation and Energy Act, Section 10202). In 2010, berries, citrus, and hops joined the network.

- Since then, NCPN-Berries has provided clean plants to state-certified programs, nurseries, and producers, raise awareness of the importance of clean plants and educate growers and nurseries on the impact of virus diseases.

- Today, the NCPN-Berries include North Carolina State University, University of Arkansas, Oregon State University/USDA-ARS Corvallis, OR, and Foundation Plant Services in California

NCPN-Berries, Corvallis, OR
Partnership with OSU Plant Clinic

- In May 2024, NCPN-Berries in Corvallis (USDA-ARS) began a restructuring initiative in collaboration with Oregon State University's Plant Clinic to expand testing capacity and better serve the industry.

- This partnership, launched in November 2024, rebranded the center as the Oregon Clean Plant Center (OCPC) and introduced a new system to serve both private and public stakeholders.

Goals

1. **Core Goal**: Optimize the production, maintenance, and distribution of clean plants
    - Maintain and distribute G1 foundation genotypes and advanced breeding selections
    - Re-test G1 genotypes and test pre-G1 material
    - Operate a CIP (Controlled Import Permit) to test and release imported genotypes
    - Eliminate target pathogens from key genera: Fragaria, Ribes, Rubus, Sambucus, and Vaccinium
    - Provide phytosanitary testing services to meet industry needs
2. **Networking & Communication**: Promote the use of clean plants through outreach and collaboration
3. **Center Optimization**: Standardize and improve systems across all berry centers
4. **Technology & Testing**: Adopt cutting-edge technologies to remain competitive and build consumer confidence
5. **Sustainability**: Ensure the long-term success of NCPN-Berries

OREGON CLEAN PLANT CENTER

Research on:

- New detection methods for detecting
- New virus cleaning methods
- More efficient positive control approaches
- Dispersion mechanisms

USDA-ARS, Corvallis OR

Public Service

HCDPMRU Virology

HCPGIRU Breeding Programs

- Maintenance of G1 (Named genotypes)-TC/SH
- Maintenance and processing of breeding Advanced selections - TC/GH
- Testing of G1 and Advanced selections by Graft indexing, ELISA, PCR
- Therapy
- Distribution of advance selections
- Prepare G1 for distributions
- Maintain positive control collection
- Keep records of legacy operations
- Database entries (Salesforce)

Oregon State University

Plant Clinic Director

Industry Service

Plant Clinic

OCPC Assistant Director

- Structure of Fee based services
- Testing for Industry Samples
- Process CIPs
- Process distribution of licensed materials
- Review and update testing protocols
- Review OCPC program operation: protocols, workflows, timelines
- Develop SOPs and Quality Standards
- Suggest operation and process improvements
- Explore alternative LIMS
- Oversee: curation and maintenance of G1 and advanced selections, positive controls collection and distribution and reports.

Mission

The Oregon Clean Plant Center (OCPC) supports the biosecurity of the U.S. berry industry by providing genotypes rigorously tested and verified to be free of targeted pathogens, maintaining a foundation of industry-important genotypes, and delivering accurate and reliable pathogen testing services.

SERVICES

Pathogen Testing: ELISA, RT-PCR, PCR, qPCR, HTS for viruses and other pathogens in *Fragaria*, *Ribes*, *Rubus*, *Sambucus*, and *Vaccinium*. Graft indexing: *Rubus* into *R. occidentalis* cv. *Munger*; *Fragaria* into *F. vesca* var. *semperflorens* 'Alpine'.

Foundation Collection: Breeders and industry partners submit genotypes for quarantine, testing, maintenance and distribution of G1 plants.

Pathogen Elimination: Infected genotypes are cleaned using microshoot tip culture and thermotherapy.

Importation of New Cultivars: Under a controlled import permit, new cultivars are imported, quarantined, tested, and released with APHIS authorization.

Distribution: provide G1 plant (cultivars and advanced selections) to nurseries or breeders as tissue culture plantlets or potted runners for Fragaria for propagation or yield trials.

SERVICES

<u>Pathogen Testing</u>:

ELISA, RT-PCR, PCR, qPCR, for viruses and other pathogens in *Fragaria*, *Ribes*, *Rubus*, *Sambucus*, and *Vaccinium*. Graft indexing: *Rubus* into *R. occidentalis* cv. *Munger*; *Fragaria* into *F. vesca* var. *semperflorens* 'Alpine'.

Graft indexing method for *Rubus* is still used to rule out Raspberry leaf curl disease and unknown viruses.

Pathogen Testing:

ELISA, RT-PCR, PCR, qPCR, for viruses and other pathogens

Host Species	Number of test available
Rubus	38
Fragaria	25
Vaccinium	22

- Stakeholders choose the test they want us to perform
- Genotypes introduced to OCPC are tested for the full panel
- Starting this year, we are running HTS on 20 Named Cultivars. The goal is to include a batch of Genotypes for HTS every funding cycle until the whole collecting had been covered.

Pathogen testing

Diagnostic tests conducted for May 2024: 8,881	
ELISA:	6,934
PCR:	1,523
Grafted genotypes:	>424

Public Service

- ✓ ELISA:

OR Rubus	5,430
OR Fragaria /Vaccinium	184
WA Rubus	1,320

- ✓ PCR: Positive control collection, advance selections round
- ✓ Grafting:

Rubus Breeders seasonal	154
Advanced Selections	119

Industry Service

Spring and Fall 2024

- ✓ Rubus grafting: 151
- ✓ Number of Companies: 4

2025 so far

- ✓ Rubus grafting: 108
- ✓ Pathogen test: 2055
- ✓ Number of Companies: 4

Foundation collection

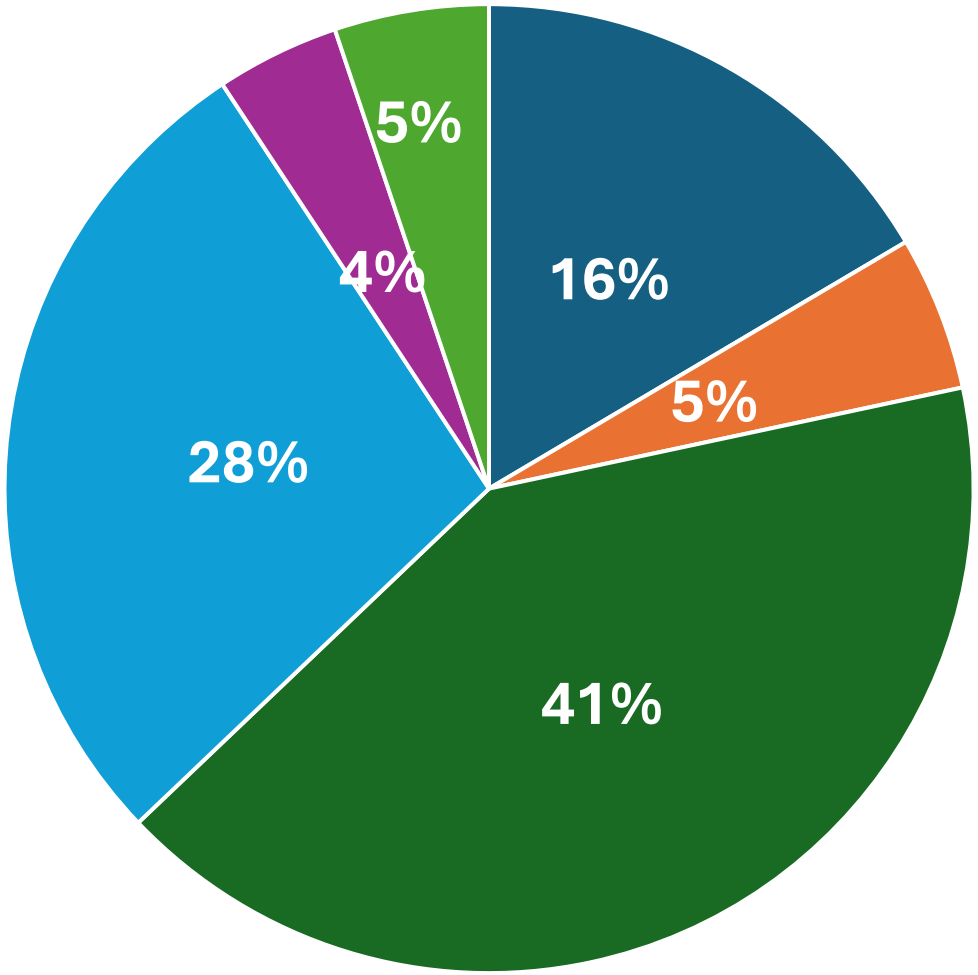
Accessions Maintained

Number of Genotypes	
Name Cultivars (G1)	97
Advanced Selections (pre-G1)	101
TOTAL	198

Named Cultivars (G1): 97	
USDA	24
WSU	10
UAK	10
MSU	1
Public Domain	49
Unknown	3

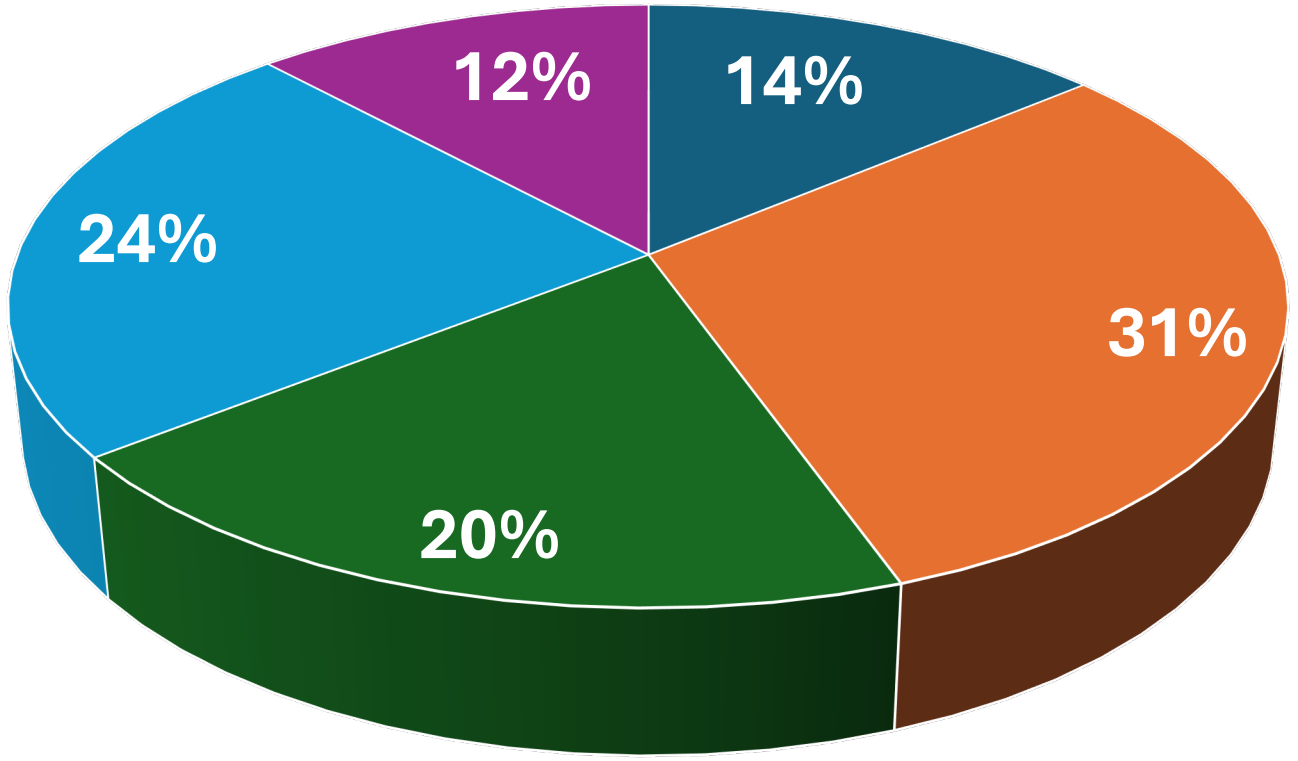
Advanced Selections: 101	
USDA	74
WSU	20
UAK	7

Named Cultivar (G1)



N=97 G1 Genotypes

Advanced Selections (101)



N=101
Advanced
selections

Strawberry
Blueberry
Blackberry
Red Raspberry
Black Raspberry

Maintenance/Pathogen testing

Virus infected positive control curate collection under APHIS permit

Virus Positive Controls Collections FY24	
Plants Total in GH5	~200

Ongoing actions:

- ✓ Update and clean database for GH5
- ✓ Continue testing to confirming presence of virus recorded
- ✓ Identify redundancies and lack-off records
- ✓ devitalization of redundancies identified – near 40 plants devitalized so far

Next:

- Institute retention policy for advanced selections: 2 years for clean up and first 3 years of maintenance cycle.
- Then the breeder will pay a maintenance fee
- Communication with each breeder to review the current inventory of genotypes under our custody

Pathogen Elimination

Infected genotypes are cleaned using microshoot tip culture and thermotherapy.

Heat therapy diagram for strawberry. Pictures from Nola Moiser.

Next:

- Try Ribavirin chemotherapy for RBDV and SCV, using protocol from CIP
- Training in cryotherapy and plan for implementation

Importation of New Cultivars

Under a controlled import permit, new cultivars are imported, quarantined, tested, and released with APHIS authorization.

FY24	
CIP accession Maintained	1 (three plants) to be released

- CIP 588 current (until 2027)
- One request for three genotypes

Distribution

Plant Distribution for FY24:	
Requested Genotypes	28
Plant unit distributed	129

2 Public programs	12 genotypes, 31 plants
6 Companies	16 genotypes, 98 plants

Positive Controls distributed for FY24:	
Positive Controls	9

Picture from Nola Mosier

Ongoing 2025

- Distribution workflow from OSU: Formats and disclaimers (Preparing distribution for South Africa)
- Recent distribution of Willamette, one of our oldest Named Cultivars

Ongoing actions

- ✓ USDA-ARS-Corvallis OR had invested more the $70.000 on improving greenhouses regulated by APHIS. A greenhouse that host positive controls and a greenhouse that host advanced selections, grafting and CIPs. Irrigation system to be installed in all benches.
- ✓ USDA-ARS-Corvallis OR had kindly contributed with $105.000 in salary and benefits for Tissue Culture and Plant Maintenance technician, and $50.000 in supplies.

- ➢ One growth chamber for quarantine and one growth chamber for thermotherapy broke. Cost to replacement would be over $35K

Ongoing actions

- Updated contact list and continued contact with stakeholders
- Fee cost structure through OSU Plant Clinic
- Developed forms for submissions and reports. Including disclaimers
- Continued review and validation of PCR and ELISA protocols currently used for *Rubus*, *Fragaria* and *Vaccinium*
- Curating inventory of G1, Advanced selections and positive control hosted by OCPC
- Reviewing workflow and timeline for OCPC operations areas
- Preparing SOPs for each operation of each area
- Draft of OCPC Operation Manual
- Participate in the ADAPT CORE trial

SOPs: Work in progress

OCPC NCPN-B | Oregon State University | USDA Agricultural Research Service

Leaf Grafting for Virus Indexing in *Rubus* Munger		
OCPC-SOP-X-v.01		Page #: 1 of 7

Purpose:
To detect viruses in *Rubus* germplasm by grafting suspect plant material onto virus-free *Rubus occidentalis* Munger, an indicator plant.

Reagents & Equipment Required:

- Virus-free *Rubus* Munger plants (indicator plants)
- Suspect donor plant material (kept cool in a cooler, with fully intact leaf petioles)
- Sterile razor blades
- *Sealtex* or equivalent grafting tape, cut into ~1" squares
- Pre-prepared labels with accurate plant IDs in advance
- Pencil for noting graft dates on pot labels
- Pot labels (weatherproof)
- 70% ethanol and 95% ethanol
- Pruners (small, precise pruners for trimming *Rubus*)
- Beaker with water (for hydrating donor tissue)
- Paper towels
- Graft log sheet
- Bucket for disposal of indicator plant tissue waste
- Hot caps (2-liter bottles with bottoms cut off)
- Two petri dishes (for sterilizing pruners and razor blades in ethanol)
- Sharps disposal container for used razor blades
- Clear plastic bags (for plants where hot caps do not fit)
- Bamboo stakes (thin, approximately 1-2 feet)
- Pruners for cutting bamboo
- Tapener tool or plant tying tape (to secure bamboo stake to plant stem)

Procedure:
Preparation—See *Figure 1* for workspace setup

1. Select healthy, actively growing *Rubus occidentalis* cv. Munger plants (3-4 weeks old) as recipient. Plants should be free of disease and uniform in size.
2. Ensure donor plants are actively growing and turgid.
3. Disinfect all tools and surfaces with 70% ethanol before beginning and between each graft.
4. Confirm that all labels match between donor plant material and recipient pot labels.
5. Lay out a pre-moistened brown paper towel to keep donor leaf material cool and hydrated during trimming and graft preparation.

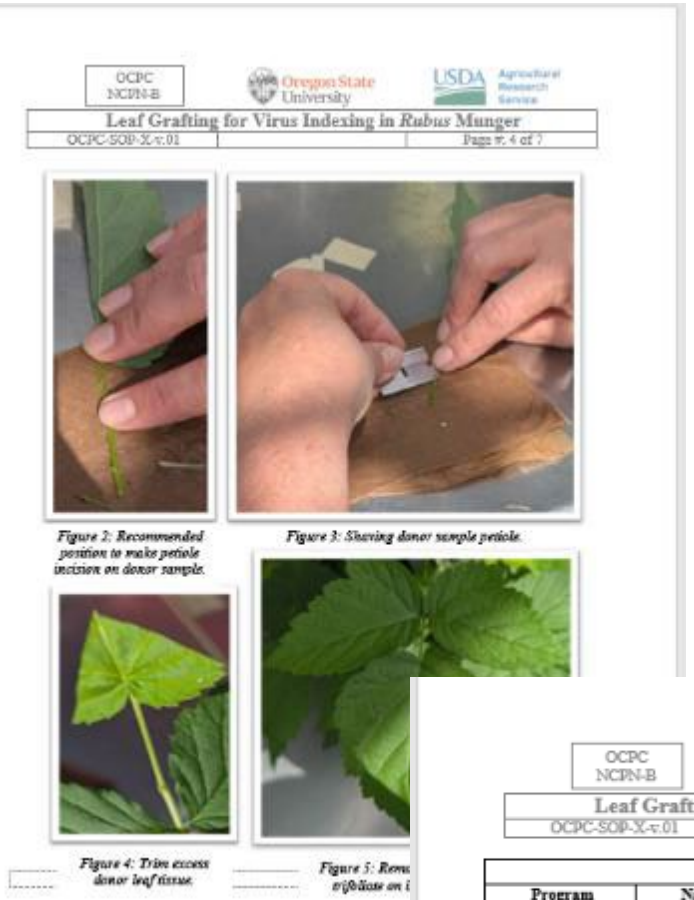


Figure 2: Recommended position to make petiole incision on donor sample.

Figure 3: Showing donor sample petiole.

Figure 4: Trim excess donor leaf tissue.

Figure 5: Remove trifoliate on i

OCPC NCPN-B | Oregon State University | USDA Agricultural Research Service

Leaf Grafting for Virus Indexing in *Rubus* Munger		
OCPC-SOP-X-v.01		Page #: 7 of 7

PI Version Approval

Program	Name	Signature	Date
USDA-ARS	Walter Mahaffee		
OSU-PC	Mana Ohkura		
OCPC	Cristian Olaya		

Revision Record

Version	Date Revised	Author	Description
Original	7/7/2025	Drafted by CoPilot, edited by Danielle Scutero, Amanda Lake, Cristian Olaya	-Draft of *Rubus* grafting for virus indexing. -Editing of *Procedure, Reagents & Equipment Required,* and *Figures.*

Oregon Clean Plant Center
(NCPN-Berries, Corvallis, OR)

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Oregon State University,
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USDA-ARS, Horticultural Crops Disease and
Pest Management Research Unit, Corvallis, OR

Start clean, stay clean
NCPN
National Clean Plant Network

Start clean, stay clean

Oregon State University

USDA HORT CROPS RESEARCH 50

Acknowledgments!

- Walt Mahaffee USDA-ARS, Corvallis, OR,
- USDA-ARS, Corvallis, OR, Staff
- B & PP Department, OSU Staff
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- ACPC Ioannis and Dan
- FPS Maher, Teresa and Staff
- Oscar Hurtado USDA APHIS PPQ PGQP
- Dr. Robert Martin

Funding

- USDA-APHIS
- USDA-ARS, Corvallis, OR (in kind contribution)
- Future check writers

Achievements & Future Developments of the NCPN Quality Initiative

NCPN Quality

A forum for exchange on best practices

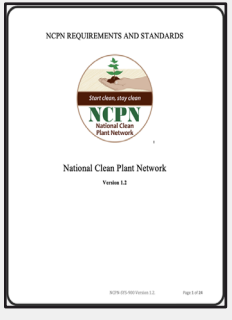
Irene Lavagi-Craddock, UCR
Fatima Osman, UCD

NCPN Quality Timeline

2019-2020

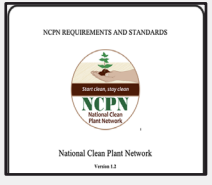
- Quality workshop; *Riverside 06/19*
- Steering Committee

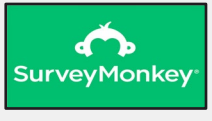
2020-2021



- NCPN Centric Standards
- QMS Training
- Auditor's workshop
- Webinar

2021-2022



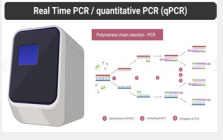


- Rolling out NCPN Centric Standards
- Quality Survey

2022-2023

- NCPN Protocols & Procedures
- Positive Control List

2023-2024



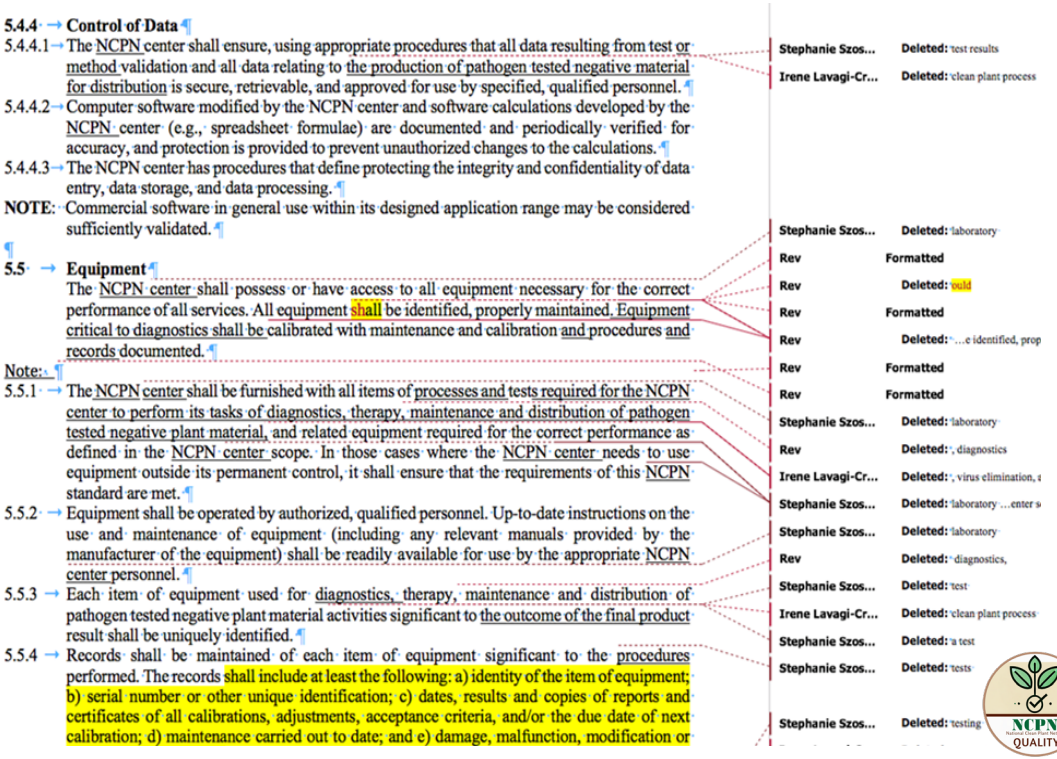
- qPCR Training
- HTS Training
- Mini Survey 2023

2024-2025

- Cross Commodity Collaboration; *Think Tank 06/24*
- QMS Refresher; *04/25*

Building NCPN-Centric Standards

- Christie Almeyda
- Kristen Aslan
- Consuelo Estevez
- Jo Foster
- Scott Harper
- Oscar Hurtado
- Lori Leong
- Bob Martin
- Ben Rosson
- Stephanie Szostek
- Karen Snover-Clift
- Dawn Dailey O'Brien
- Kathy Burch
- Erich Rudyj
- David Prokrym
- Patrick Shiel
- Sarah Trujillo
- Irene Lavagi-Craddock
- Fatima Osman



5.4.4 → Control of Data ¶

5.4.4.1 → The NCPN center shall ensure, using appropriate procedures that all data resulting from test or method validation and all data relating to the production of pathogen tested negative material for distribution is secure, retrievable, and approved for use by specified, qualified personnel. ¶

5.4.4.2 → Computer software modified by the NCPN center and software calculations developed by the NCPN center (e.g., spreadsheet formulae) are documented and periodically verified for accuracy, and protection is provided to prevent unauthorized changes to the calculations. ¶

5.4.4.3 → The NCPN center has procedures that define protecting the integrity and confidentiality of data entry, data storage, and data processing. ¶

NOTE: Commercial software in general use within its designed application range may be considered sufficiently validated. ¶

¶

5.5 → Equipment ¶

The NCPN center shall possess or have access to all equipment necessary for the correct performance of all services. All equipment shall be identified, properly maintained. Equipment critical to diagnostics shall be calibrated with maintenance and calibration and procedures and records documented. ¶

Note: ¶

5.5.1 → The NCPN center shall be furnished with all items of processes and tests required for the NCPN center to perform its tasks of diagnostics, therapy, maintenance and distribution of pathogen tested negative plant material, and related equipment required for the correct performance as defined in the NCPN center scope. In those cases where the NCPN center needs to use equipment outside its permanent control, it shall ensure that the requirements of this NCPN standard are met. ¶

5.5.2 → Equipment shall be operated by authorized, qualified personnel. Up-to-date instructions on the use and maintenance of equipment (including any relevant manuals provided by the manufacturer of the equipment) shall be readily available for use by the appropriate NCPN center personnel. ¶

5.5.3 → Each item of equipment used for diagnostics, therapy, maintenance and distribution of pathogen tested negative plant material activities significant to the outcome of the final product result shall be uniquely identified. ¶

5.5.4 → Records shall be maintained of each item of equipment significant to the procedures performed. The records shall include at least the following: a) identity of the item of equipment; b) serial number or other unique identification; c) dates, results and copies of reports and certificates of all calibrations, adjustments, acceptance criteria, and/or the due date of next calibration; d) maintenance carried out to date; and e) damage, malfunction, modification or

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Irene Lavagi-Cr... **Deleted:** clean plant process

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Stephanie Szos... **Deleted:** test

Irene Lavagi-Cr... **Deleted:** clean plant process

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NCPN Requirements & Standards

1.0 MISSION

2.0 ADMINISTRATIVE REQUIREMENTS

2.1 Organization, Management and Personnel
2.2 Finance and Budget

3.0 ACCREDITATION PROCESS

4.0 MANAGEMENT REQUIREMENTS

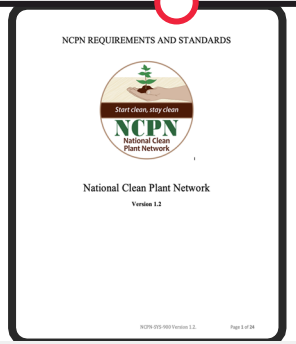
4.1 Organization and Management
4.2 Quality System
4.3 Document Control
4.4 Review of Request or Agreement
4.5 Subcontracting of Test Services
4.6 Purchasing Services and Supplies
4.7 Customer Feedback
4.8 Control of Non-conforming Testing and Test Results

4.9 Corrective and Preventative Actions
4.10 Records
4.11 Internal Audits
4.12 Management Reviews

5.0 TECHNICAL REQUIREMENTS

5.1 General
5.2 Personnel

Set of requirements to help formulate coordinated activities directing the quality of the product.


NCPN REQUIREMENTS AND STANDARDS

Start clean, stay clean

NCPN National Clean Plant Network

National Clean Plant Network

Version 1.2

5.0 TECHNICAL REQUIREMENTS

5.7 Samples and Sample Handling

5.7.2 The NCPN center shall outline the procedures for the receipt, handling, protection, storage, retention, disposal, and internal and external transfers of plant material including provisions necessary to protect the integrity of the plant material, and the interests of the NCPN center and the client/stakeholder.

5.7.3 The NCPN center shall have a system for identifying plant material that ensures no confusion between original plant material and the final product. The identification shall be retained throughout the life of the plant material and its derived final product in the NCPN center and linked to the report.

NCPN QUALITY

NCPN Quality Management

The 4 Core Steps of QMS

Say What You Do

Clearly document your processes

Do What You Say

Ensure adherence to documented processes across the organization.

Prove It

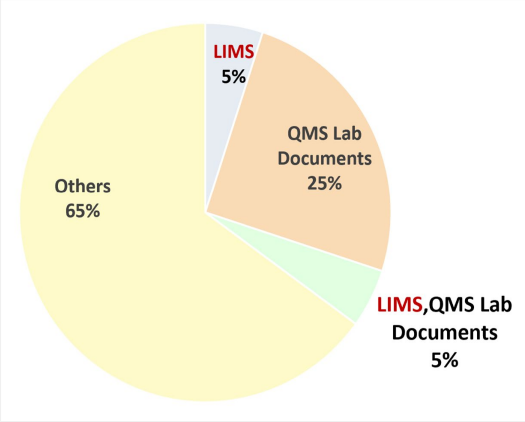
Provide evidence of compliance through records

Improve It

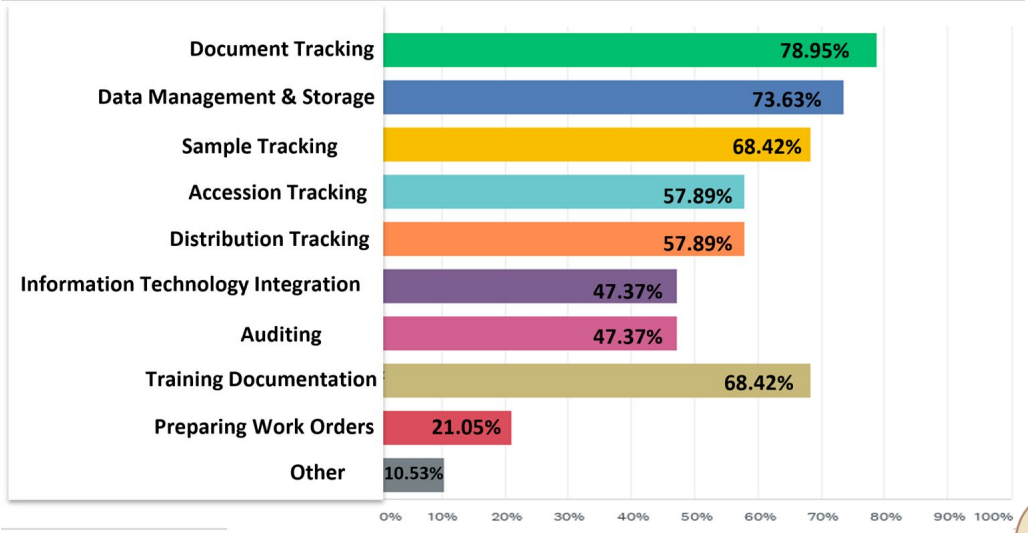
Continuously seek opportunities for ongoing improvement.

Survey Insights

Q63: What have your Centers/Programs done so far to adopt QMS?



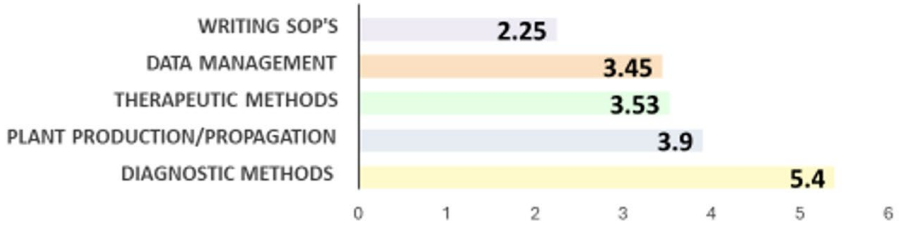
Q: Please mark the areas where your Center/Program could most benefit from quality principles.

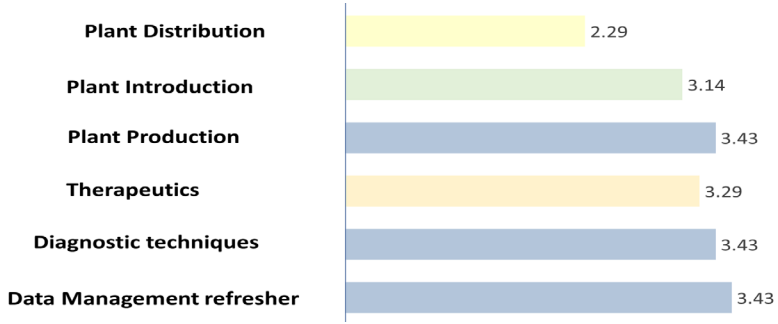


Survey Insights- Training

- qPCR
- High Throughput Sequencing
- SOPs
- Introduction/ Distribution
- Data Management Platforms
- Virus Elimination Techniques

Q44: Please rank the following areas of training in order of importance to your program?



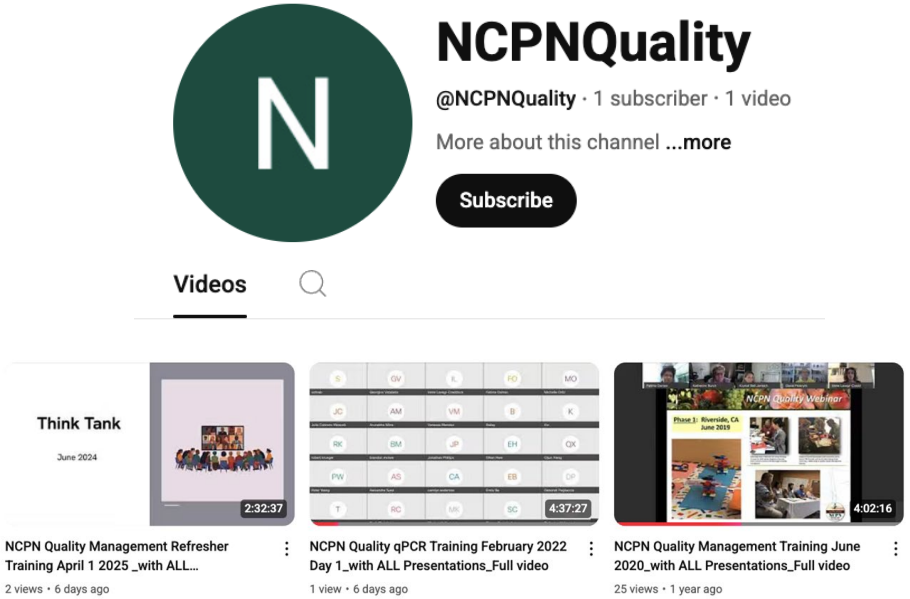


Training

qPCR

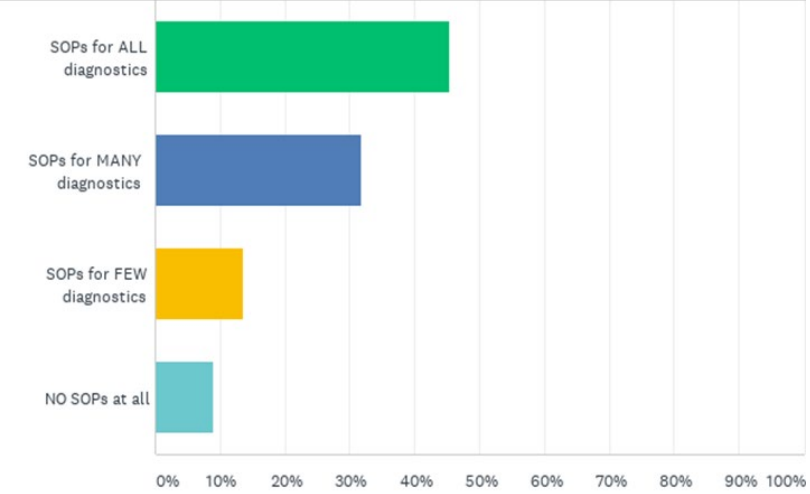
HTS

QMS Refresher

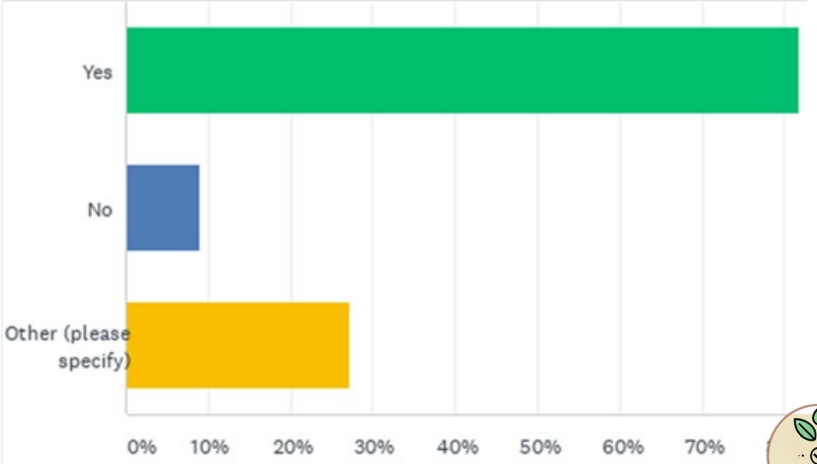


Sharing the Toolbox Across the Network

Q40: Do you have SOPs for diagnostics techniques performed at your center?



Q42: Would you be willing to share positive controls with other Centers?



Positive Controls

Citrus Sharing Positive Controls

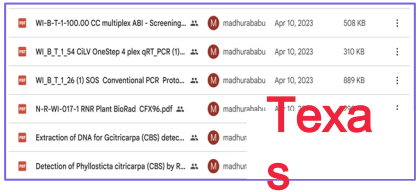
NCPN-Citrus Positive Control List and Contacts

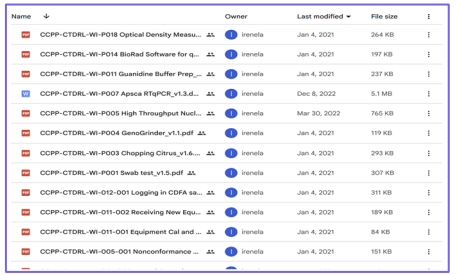
NCPN-Citrus Centers and Emails

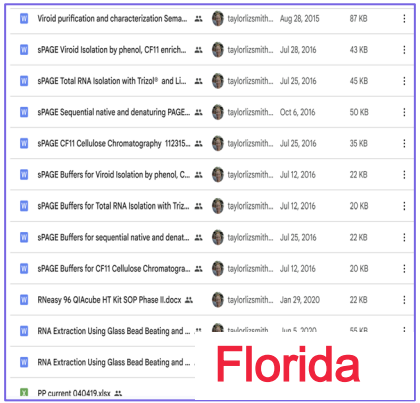
# +ve con.	Pathogen	NCPN-CA		NCPN-Louisiana	NCPN-Maryland	NCPN-Florida	NCPN-Puerto Rico	NCPN-Texas	NCPN-Alabama	NCPN-Hawaii	NCPN-Arizona
		CA-CCPP	CA-NCGRCD	Louisiana State U.	EPCC	FDACS (CGIP)	University of Puerto Rico	Texas A & M University	Auburn University	University of Hawaii	
Viruses											
1	Citrus Tatter leaf virus (syn. Apple stem grooving virus) (CTLV)	Yes, in planta	Yes, in planta			Yes, in planta		Yes, in planta			No Positive Controls
2	Citrus vein enation virus (CVEV)	Yes, in planta	Yes, in planta			Yes, Nucleic acid				Yes, in planta	
3	Citrus yellow vein associated virus (CYVaV)	Yes, in planta	Yes (?), in planta								
4	Citrus sudden death-associated virus (CSDaV)										
5	Citrus jingmen-like virus										
6	Citrus virga-like virus (CVLV)										
7	Satsuma dwarf virus (SDV)		Yes (?), in planta								
8	Citrus mosaic badnavirus (CiMV)/Citrus yellow mosaic virus(CYMV)				Yes (?)						
9	Natsudaidai dwarf virus (NDV)										
10	Navel infectious mottling virus (NIMV)										
11	Hyuganatsu virus										
12	Citrus chlorotic dwarf-associated virus (CCDaV)		Yes, in planta		Yes						
13	Citrus yellow mosaic virus(CYMV)/Citrus mosaic badnavirus (CiMV)		Yes, in planta		Yes (?)						
14	Citrus yellow vein clearing virus-Mandarivirus (CYVCV.2)	Yes, Nucleic acid			Yes			Yes, nucleic acid			
15	Citrus leprosis cilevirus C (CiLV-C)		Yes (?), Nucleic acid		Frozen tissues and total nucleic acid			Yes, nucleic acid		Yes, nucleic acid	
16	Citrus leprosis cilevirus C2 (CiLV-C2)				Frozen tissues and total nucleic acid					Yes, in planta	
17	Citrus leprosis dichorhavirus N (CiLV-N)				Total Nucleic acid						
18	Citrus chlorotic spot virus (CiCSV)				Total Nucleic acid						
19	Orchid fleck dichorhavirus (OFV)				Frozen tissues and total nucleic acid					Yes, in planta	
20	Citrus variegation illarvirus (CVV)=Citrus crinkly leaf virus	Yes, in planta	Yes, in planta			Yes, in planta					
21	Citrus leaf rugose ilarvirus virus(CLRV)		Yes, in planta			Yes, in planta					
22	Citrus chlorotic dwarf-associated virus (CCDaV)		Yes, in planta		Yes						
23	Citrus virus A	Yes, in planta	Yes, in planta			Yes, in planta		Yes, in planta			
24	Citrus Concave gum associated virus		?			Yes, in planta		Yes, in planta			
25	Citrus Leaf blotch virus 2		?								
26	Citrus Tristeza virus (CTV)	Yes, in planta	Yes, in planta		Yes	Yes, in planta (Mild & Severe)		Yes, in planta		Yes, in planta	
27	Citrus psorosis virus(CPsV)	Yes, in planta	Yes, in planta			Yes, in planta		Yes, in planta			
28	Citrus leaf blotch virus (CLBV) syn. Dweet mottle virus	Yes, in planta	Yes, in planta			Yes, in planta	Yes, Nucleic acid	Yes, nucleic acid			

Protocols & Procedures

Citrus Harmonization Efforts







Think Tank Outcomes: Our Shared Priorities

Expand Training

Priorities in High-Throughput Sequencing (HTS), SOP development, and practical skills

"We should have our training initiatives like next-gen sequencing that will help us to move forward."

Strengthen Data Management

Explore adaptable LIMS platforms, record digitization, smarter data sharing.

Enhance Collaboration

Cross-commodity workshops, in-person exchanges, online resources, tailored support visits.

"Just hearing from other centers [...] can bring up issues and opportunities we might not have considered otherwise."

QMS Refresher: Lessons Learned

Living Framework

Continuous improvement, collective engagement, shared responsibility.

Practical Training

SOP development and resource-sharing platforms to strengthen daily practices

Smarter Records

Strong interest in digitization and adaptable record systems

Key Achievements & Opportunities

Key Achievements

- 📈 Training in diagnostics (qPCR, HTS)
- 📝 Training in Quality Management
- 📜 Developed Quality Standards
- 🔄 Shared protocols and procedures
- 🔔 Continuous community feedback
- 🤝 Collaborated with other initiatives

Opportunities

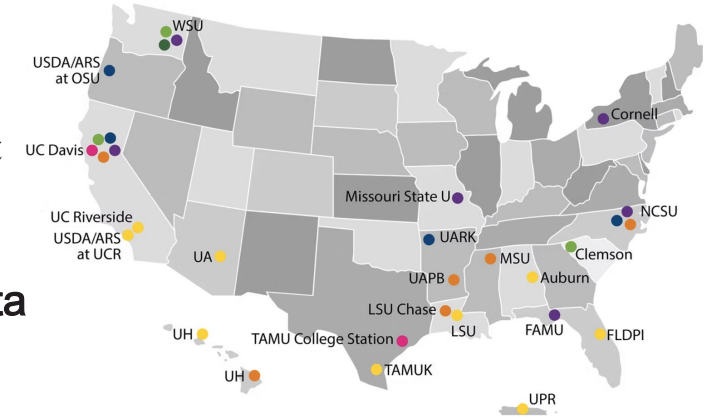
- Embed QMS & SOP training in daily operations
- Advance digitization and adaptable record systems
- Expand cross-commodity collaboration & exchanges
- Build shared online training portal
- Develop criteria for voluntary quality reviews

Implementation

From Ideas to Action

What do *YOU WANT?*

- What **training formats** would help your staff most (online sessions, full workshops with an in-person component, self-paced modules)?
- How can we make QMS tools and SOP development more practical for your center?
- What features would you need in a **shared LIMS/data management** portal?



- Where do you see the biggest opportunities for **cross-commodity collaboration** ?
- What support (visits, mentoring, online sharing) would have the most impact?

A Summary of NCPN Economic Working Group Studies

September 10, 2025

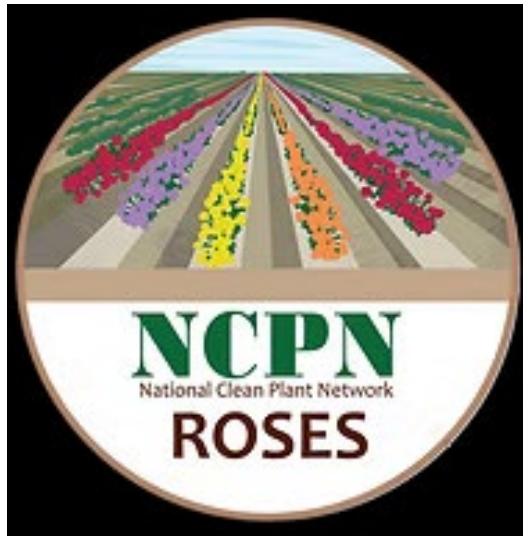
Kristen Park
Dyson School of Applied Economics and Management
Cornell University

- Yeh, A., Park, K. S., Gómez , M. I., and Fuchs, M. (2019). *A Review of Economic Studies on Pathogen-Tested Plant Materials and Clean Plant Programs for Specialty Crops.* Working Paper WP 2019-12. Charles H. Dyson School of Applied Economics and Management, Cornell University.
- "Economic Studies Reinforce Efforts to Safeguard Specialty Crops in the United States." Authors M. Fuchs, C. V. Almeyda, M. Al Rwahnih, S. S. Atallah, E. J. Cieniewicz, K. Farrar, W. R. Foote, D. A. Golino, M. I. Gómez, S. J. Harper, M. K. Kelly, R. R. Martin, T. Martinson, F. M. Osman, K. Park, V. Scharlau, R. Smith, I. E. Tzanetakis, G. Vidalakis, and R. Welliver. Published Online: 29 October 2020.

Economic Benefits of Using Clean Plant Materials

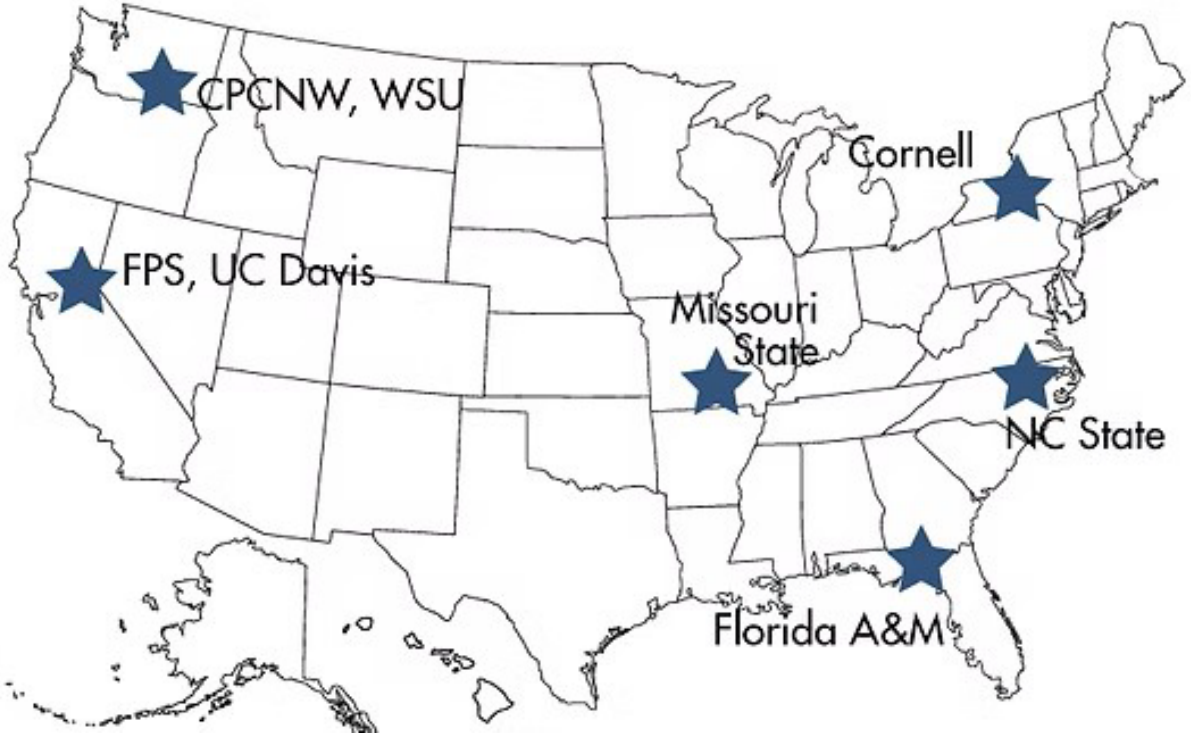
Economic benefits of using certified clean hops

The relationships between generation, virus loads, and yield in us sweetpotatoes (continuing)



Assessing the socioeconomic impact of rose rosette disease management approaches

Benefit Cost Ratios for NCPN Centers

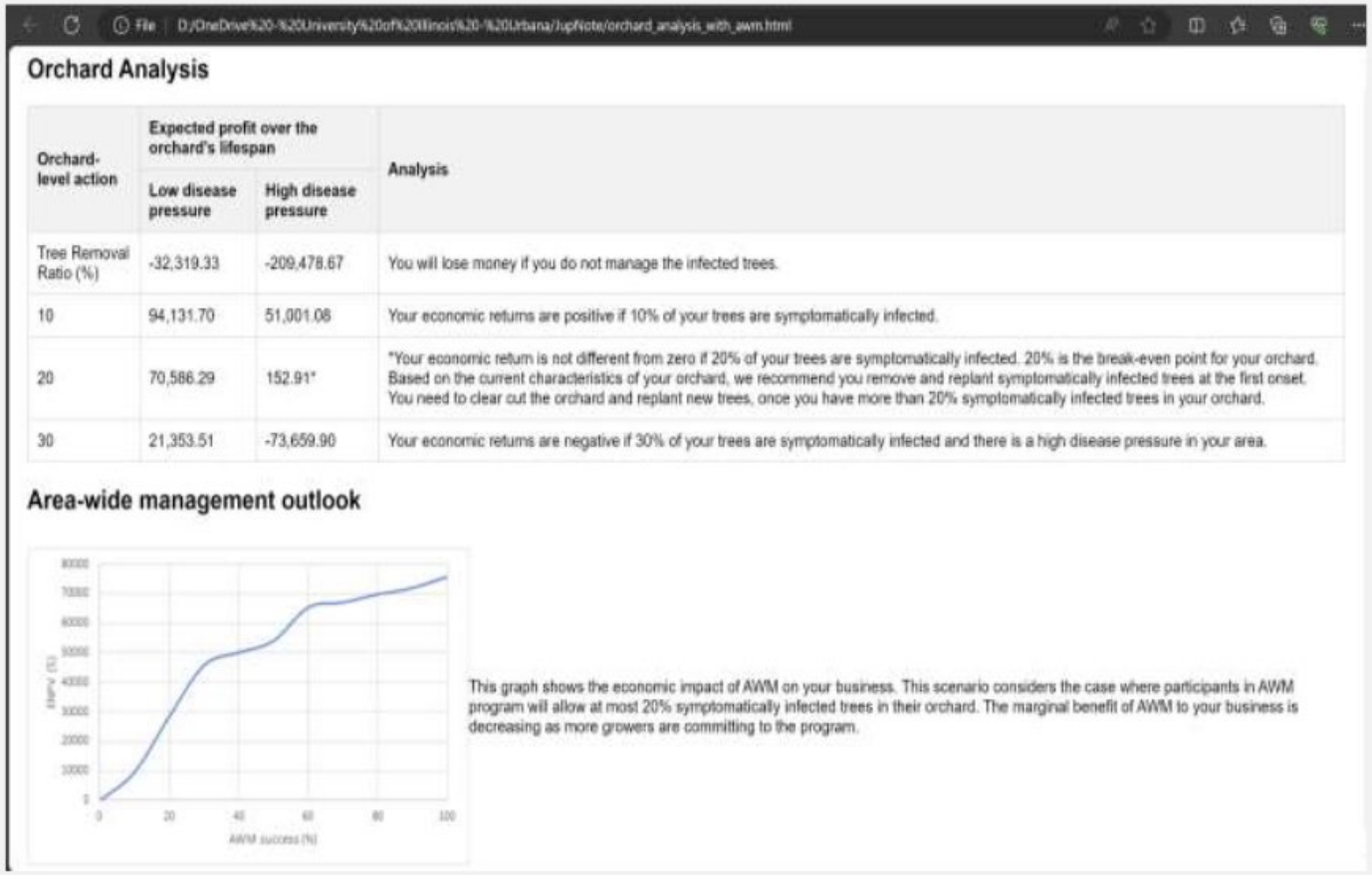


Foundation Plant Services in CA, "Returns to public investments in clean plant centers"

Grapes – Eastern NCPN-Grapes Center (underway)

Citrus – Citrus Clonal Protection Program in CA (underway)

POTENTIAL DECISION AID TOOL TO CONTROL WESTERN X-DISEASE



Orchard Analysis

Orchard-level action	Expected profit over the orchard's lifespan		Analysis
	Low disease pressure	High disease pressure	
Tree Removal Ratio (%)	-32,319.33	-209,478.67	You will lose money if you do not manage the infected trees.
10	94,131.70	51,001.08	Your economic returns are positive if 10% of your trees are symptomatically infected.
20	70,586.29	152.91*	*Your economic return is not different from zero if 20% of your trees are symptomatically infected. 20% is the break-even point for your orchard. Based on the current characteristics of your orchard, we recommend you remove and replant symptomatically infected trees at the first onset. You need to clear out the orchard and replant new trees, once you have more than 20% symptomatically infected trees in your orchard.
30	21,353.51	-73,659.90	Your economic returns are negative if 30% of your trees are symptomatically infected and there is a high disease pressure in your area.

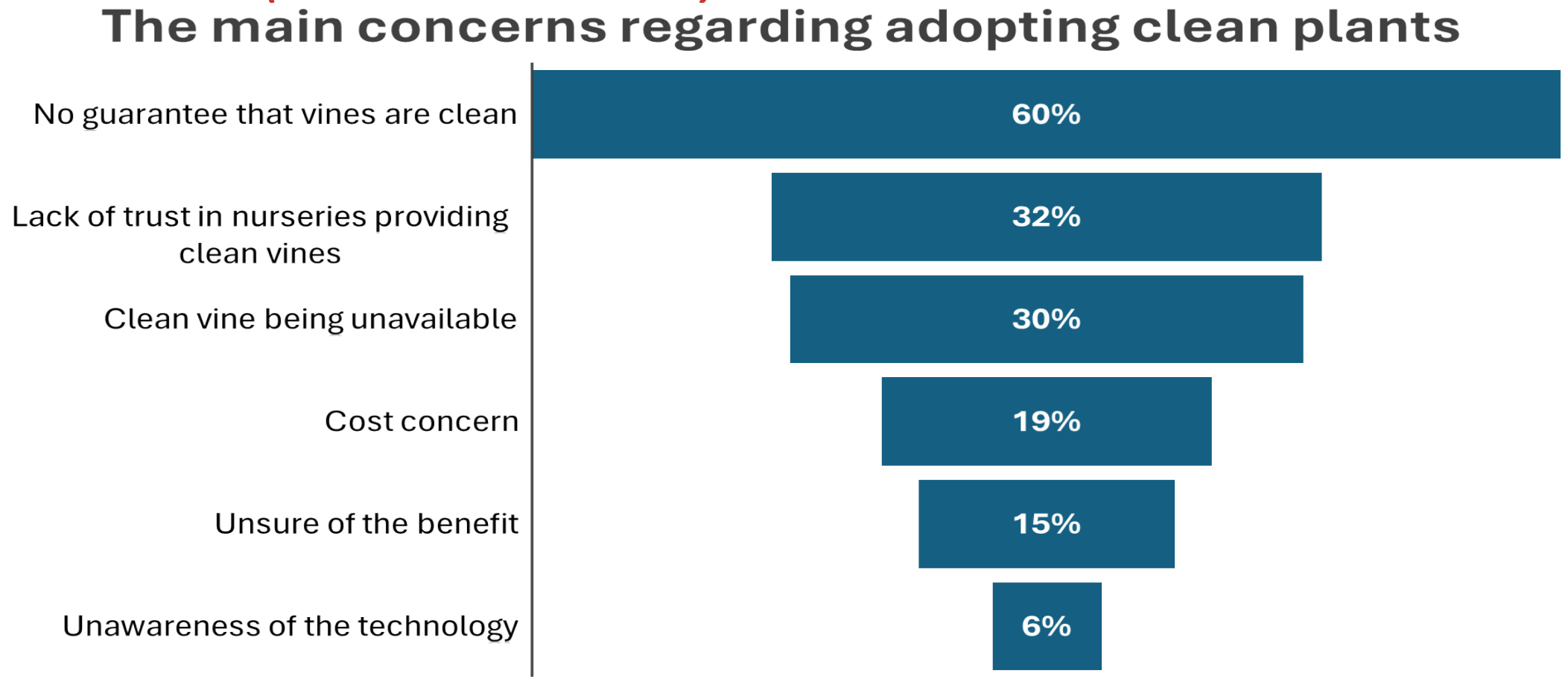
Area-wide management outlook

This graph shows the economic impact of AWM on your business. This scenario considers the case where participants in AWM program will allow at most 20% symptomatically infected trees in their orchard. The marginal benefit of AWM to your business is decreasing as more growers are committing to the program.

Figure 2: The Second Section of the Online Platform.

- Web-based platform:
  - customized research-based recommendation on the optimal tree removal rates
  - participants commit to share factual information about the number of removed diseased trees in their orchard
  - shows the potential farm-level effects of successful area-wide management.

FACTORS INFLUENCING GRAPE GROWERS' ADOPTION OF CLEAN PLANT MATERIALS – *(REPORT DRAFTED)*



Challenges and Opportunities

- What can 5 years of studies inform us about the challenges and opportunities for clean plant economic research?
  - Reliance on data
    - high quality data
    - industry data
  - Cost of getting the data
  - Industry trust and collaboration
  - Communicating research results to the industry
    - product, place, price, promotion

FUCHS, ET AL.

- *"Similarly, better understanding of socioeconomic elements that drive growers' decisions to use or not to use certified planting material would eventually provide new opportunities to strategically increase the adoption of plants derived from G1 stocks. Finally, future research efforts should address how to best communicate findings of economic studies to growers and nurseries to foster the use of clean planting material, as elegantly documented for the adoption of integrated pest management (Sherman and Gent 2014)."*

National Clean Plant Network Education and Outreach







Jennifer Nicholson
NCPN Coordinator
USDA-APHIS-PPQ

Education/Outreach History

- Since program's beginning the importance of outreach to communicate the value of clean plants has been recognized
- Crop and center-based outreach with customers and industry is the primary component, but there was an early network need for standardization and centralization of communication of materials
- NCPN E/O began in 2010 as a working group focused on website, art, and logos
- 2015-2021 Large cross network working group coordinated by 2-3 part-time co-chairs to complete outreach objectives
- 2022-present Reorganized with a smaller working group with crop coordinators and additional representation from each crop group

NCPN E/O Goals

- Promote awareness of the NCPN program with stakeholders
- Communicate the impact and value of the overall program for the specialty crop industry
- Support crop efforts to increase the use of clean plant materials by nurseries and growers

Scope of activities:

- Assess national outreach and communication priorities
- Develop and maintain national level communication products, including the NCPN website
- Coordinate information to support development of crop-specific communication products

Website: www.nationalcleanplantnetwork.org

WELCOME

The National Clean Plant Network (NCPN) is comprised of clean plant centers, scientists, educators, state and federal regulators, large and small nurseries, and growers of specialty crops that work together to ensure that plant propagation material is clean and available.

The Network was created to protect U.S. specialty crops such as berries, citrus, fruit trees, grapes, hops, roses and sweetpotatoes from the spread of economically harmful plant pests and diseases. NCPN ensures the global competitiveness of U.S. specialty crop producers by creating high standards for our clean plant programs.

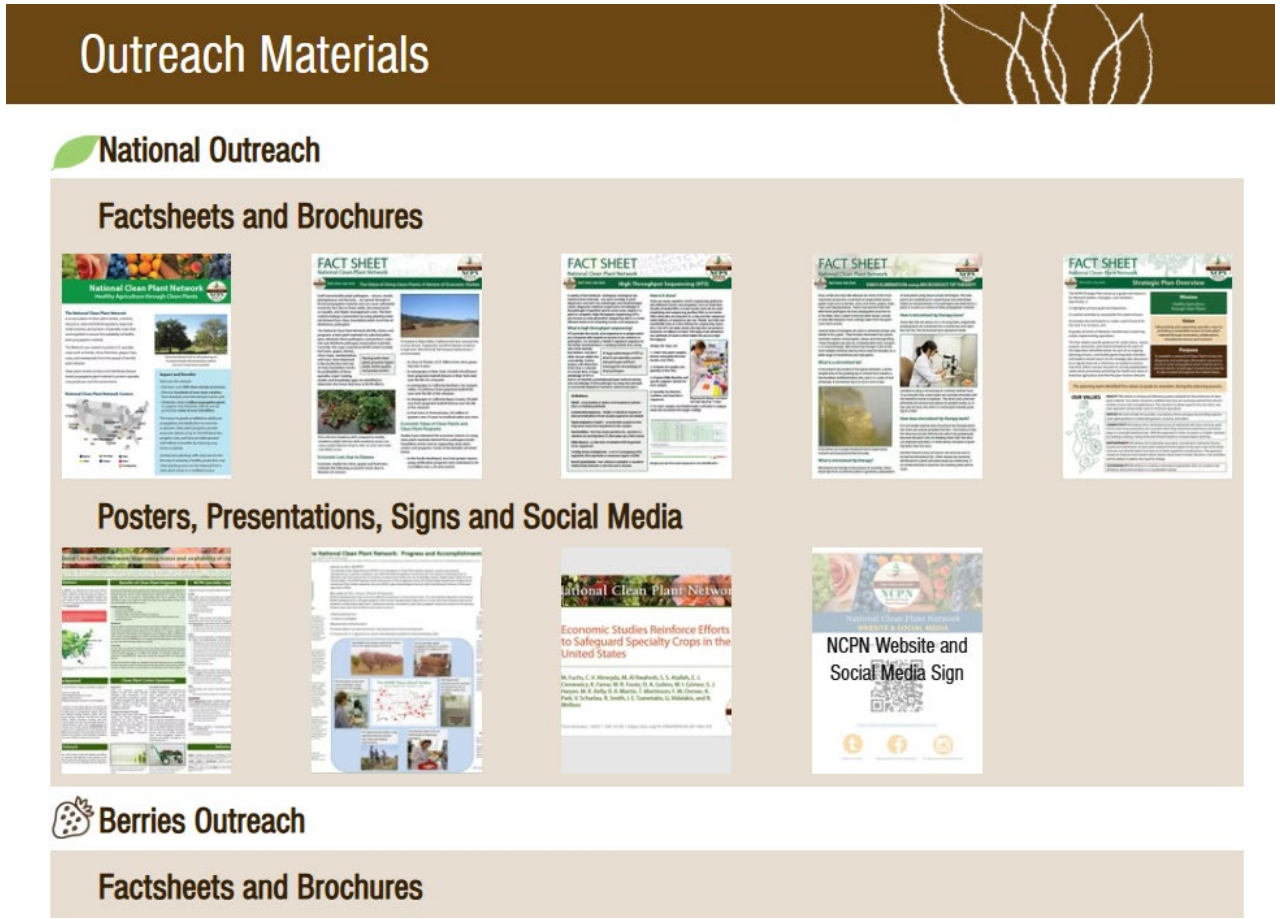
Crop Brochures and Factsheets

Brochures for each crop and national network

21 factsheets on clean plants, economic value, and key disease issues

Additional products

- Industry Testimonials
- Booth display
- Presentations overview and templates
- Resource Library of publications



Network updates

- Quarterly newsletter with over 700 recipients highlighting program and scientific developments
- Posts on website and on social media

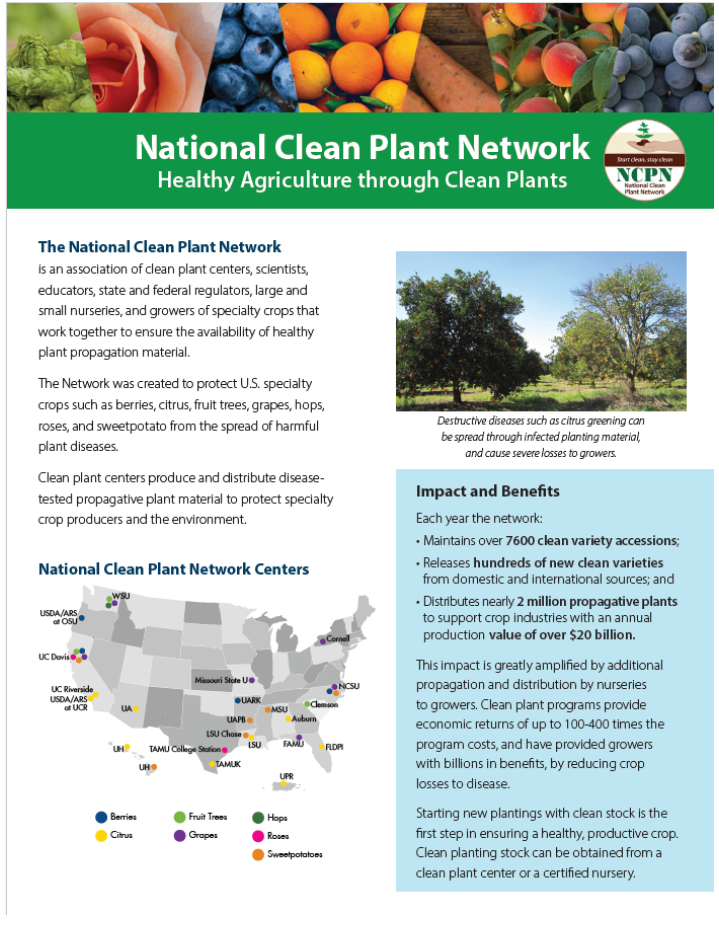
NETWORK NEWS

NCPN-Berries Keeps on Growing!

The Berries network continues to make strides in expanding and improving on services offered.

Recent Updates

- Crop websites updated
- New graphics, flyer and overview presentation



National Clean Plant Network
Healthy Agriculture through Clean Plants

The National Clean Plant Network
is an association of clean plant centers, scientists, educators, state and federal regulators, large and small nurseries, and growers of specialty crops that work together to ensure the availability of healthy plant propagation material.

The Network was created to protect U.S. specialty crops such as berries, citrus, fruit trees, grapes, hops, roses, and sweetpotato from the spread of harmful plant diseases.

Clean plant centers produce and distribute disease-tested propagative plant material to protect specialty crop producers and the environment.

National Clean Plant Network Centers

Destructive diseases such as citrus greening can be spread through infected planting material, and cause severe losses to growers.

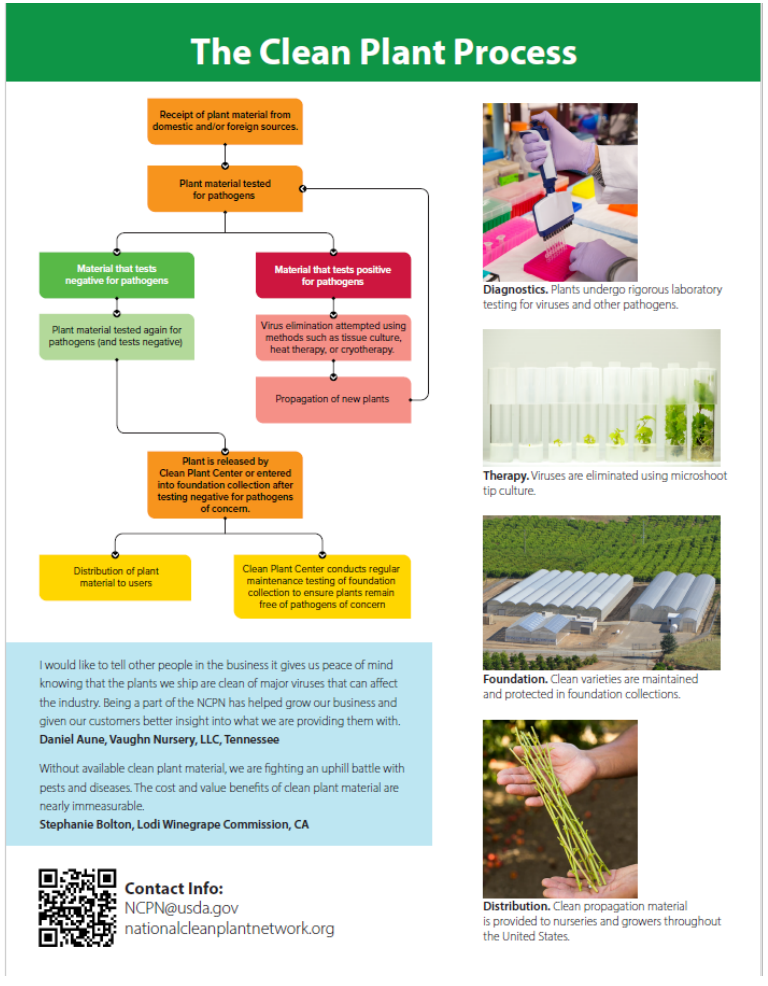
Impact and Benefits

Each year the network:

- Maintains over **7600 clean variety accessions**;
- Releases **hundreds of new clean varieties** from domestic and international sources; and
- Distributes nearly **2 million propagative plants** to support crop industries with an annual production **value of over $20 billion.**

This impact is greatly amplified by additional propagation and distribution by nurseries to growers. Clean plant programs provide economic returns of up to 100-400 times the program costs, and have provided growers with billions in benefits, by reducing crop losses to disease.

Starting new plantings with clean stock is the first step in ensuring a healthy, productive crop. Clean planting stock can be obtained from a clean plant center or a certified nursery.



The Clean Plant Process

I would like to tell other people in the business it gives us peace of mind knowing that the plants we ship are clean of major viruses that can affect the industry. Being a part of the NCPN has helped grow our business and given our customers better insight into what we are providing them with.
Daniel Aune, Vaughn Nursery, LLC, Tennessee

Without available clean plant material, we are fighting an uphill battle with pests and diseases. The cost and value benefits of clean plant material are nearly immeasurable.
Stephanie Bolton, Lodi Winegrape Commission, CA

Contact Info:
NCPN@usda.gov
nationalcleanplantnetwork.org

Diagnostics. Plants undergo rigorous laboratory testing for viruses and other pathogens.

Therapy. Viruses are eliminated using microshoot tip culture.

Foundation. Clean varieties are maintained and protected in foundation collections.

Distribution. Clean propagation material is provided to nurseries and growers throughout the United States.

Clean grape videos

- Collaboration with Penn State, Cornell, FPS
- Short videos on:
  - Why are some Grapevine Diseases not tested for in the certification process?
  - Process from grapevine entry to, and distribution from, a clean plant center
  - Why Grapevine Certification Matters?
  - What Is a “Clean Grapevine” (and how does this differ from a “certified vine”?)?
  - Grape grower and vineyard nursery owner testimonial about grapevine certification

VIDEOS

Answers from the Vineyard, Winery, and Tasting Room

By: Cain Hickey, Molly Kelly, Michela Centinari, Ph.D., Bryan Hed, Meredith Persico

A video series to answer questions that are relevant to the grape and wine industry in Pennsylvania and eastern United States.

Education/Outreach looking forward

- Original goals for the national working group largely met
- Crop strategic plans have a communication component and was identified as a priority
- Discussion topic – role of national working group coordination vs. center and crop group coordination of outreach goals

National Clean Plant Network Strategic Planning







Jennifer Nicholson
NCPN Coordinator
USDA-APHIS-PPQ

Strategic Planning Initiative

- NCPN 2021-2025 Strategic Plan and Program Implementation Guide
- Team of 10 representatives revisiting NCPN mission, vision, strategies, goals, and objectives
- Goals provide priorities for crop group and proposal planning
  - Goal 1: Network Program Operations
  - Goal 2: Advancing Special Initiatives
  - Goal 3: Governance and Networking

Mission

Healthy Agriculture through Clean Plants

Vision

Safeguarding and supporting specialty crops by providing a sustainable source of clean plant material through innovation, collaboration, translational science and outreach.

Purpose

To establish a network of Clean Plant Centers for diagnostic and pathogen elimination services to produce clean propagative plant material and to maintain blocks of pathogen-tested plant material in sites located throughout the United States.

Goal 1- Network Program Operations

Optimize the Production, Maintenance and Distribution of Clean Plants

1. **Stakeholder Driven Clean Plant Centers:** Develop a network of centers that is focused on the stakeholder and their needs for clean plant material.
2. **Existing and New Centers:** Optimize the Number of NCPN Centers to ensure for optimal and complete specialty crop coverage.
3. **Protocols, Standards, Collaboration, and Coordination:** Improvement and coordination of cleanup activities and maintenance of clean plants.
4. **Foundation Management:** Assess, support and manage a Network of nationally focused clean plant foundations and collections.
5. **Plant Availability:** Develop and maintain an up-to-date list(s) of available cultivars. Facilitate the distribution of clean products at Clean Plant Centers.
6. **Permitting:** Facilitate permits in support of Network activities.
7. **International Clean Plant Program Connections**: Centers are well connected internationally for material access and regulatory purposes.

Goal 2 – Advancing Special Initiatives

Optimize the adaptation and implementation of novel technologies and new ideas while increasing the awareness of the importance, availability, and use of clean plants

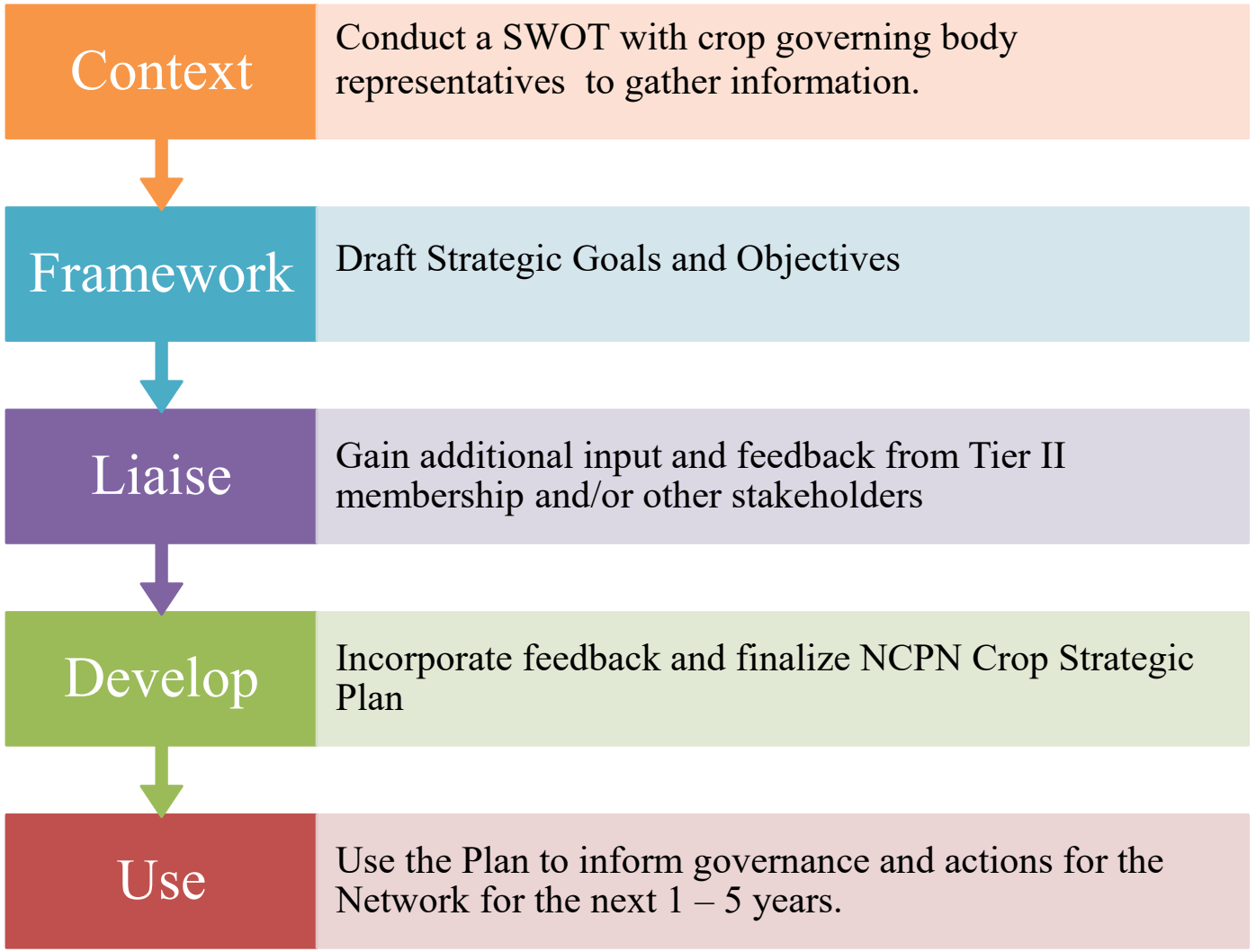
1. **Use Advanced Diagnostics and Ascertaining Risks of Not Engaging in New Technologies :** Use more sensitive and comprehensive detection methods to accelerate the production of clean plants and identify risks of not adopting new technologies.
2. **Foundation Protection:** Use new technologies to safeguard and back-up foundation material.
3. **Natural Disaster Preparation:** Secure clean material in case of natural disasters or disease outbreaks.
4. **Clean Plant Material Demand:** Determine Industry Needs for Clean Plant Material.
5. **Clean Plant Outreach:** Facilitate the adoption of clean plants by nurseries and growers through extension and outreach activities.
6. **Economics:** Assess the economic impact of the clean plant programs.
7. **Quality Assurance and Quality Control:** Establish formalized programs and processes for program Quality Management.
8. **Scientific Information Development, Use, and Sharing:** Develop agreement and establish procedures for data and information development and sharing, both internally and externally.
9. **Interface with Nursery Clean Plant Programs:** Facilitate communication among regulators, research scientists, industry, and centers to develop clean plant nursery programs to increase the use of clean plant material.

Goal 3: Governance and Networking

Optimize Network resources

1. **NCPN Program Governance:** Assess and ascertain NCPN Governance and Administration needs for FY 2021-2025.
2. **Governing Bodies:** Accommodate governance for new, small, or 'occasional' specialty crops.
3. **NCPN Program Scope:** Revisit and re-circumscribe the scope and parameters of the Network.
4. **Clean Plant Specialty Crop Business Plans:** NCPN Specialty Crop Groups and Clean Plant Centers Develop Business Plans to insure for the long-term financial sustainability of specific crops within the network.
5. **NCPN Planning Harmonization Strategy:** In developing and implementing national, crop specific, center oriented, and special topics plans, NCPN managers will strive to coordinate and harmonize all plans.
6. **Funding Stability:** Identify complementary sources of funding to support and/or enhance clean plant programs.
7. **Succession Planning:** Develop Succession plans for human resources; and including plants people, and infrastructure.
8. **Program Networking:** Governance bodies should meet regularly with stakeholders and networking groups for continuing collaboration and to engage in critical discussions around pertinent network/commodity topics.

Crop Strategic Planning Process



Grapes Strategic Plan

- **GOAL 1: Security**—Ensure virus-tested G1 stock is accessible in a timely manner
- **GOAL 2: Redundancy**—Keep two plants per cultivar, clone, selection, or rootstock of G1 stock
- **GOAL 3: Capacity**—Maintain two G1 vines of all cultivars and rootstocks of interest at two different locations
- **GOAL 4: Communication**—Virus-tested G1 material is available
- **GOAL 5: Sustainability**—Within 10 years, ensure that plants, protocols, programs, and production have infrastructure to endure

Berries Strategic Plan

- ***Goal 1: Networking/ Communication:*** Promote usage of clean plants through networking and communication.
- ***Goal 2: Optimize and Integrate Berries' Centers:*** Improve, standardize, and implement systems across centers
- ***Goal 3: Technology and Testing:*** Use best technologies to remain competitive and inspire consumer confidence
- ***Goal 4: Sustainability:*** Ensuring the future success of NCPN berries

Hops

Goal 1 Biological Security: *NCPN-Hops will provide safe sources of selections from private and public programs.*

Goal 2: Service Capacity *NCPN-Hops will establish and maintain foundation mother plants to provide clean planting stock to industry within prescribed state and federal certification schemes.*

Goal 3: Outreach *NCPN-Hops will establish and coordinate working relationships with entities certifying plants for planting and develop best management practices used by industry.*

Roses

- Goal 1: Ensure the sustainability of clean rose programs while maintaining the long term availability of priority varieties
- Goal 2: Develop and recommend guidelines for maintaining clean rose plants to extend the value of clean plant material throughout the rose production process.
- Goal 3: Effectively communicate the benefits of clean plant material and the work of NCPN

Citrus (draft)

- Improve Operational Efficiency and Effectiveness
- Enhance Consumer Education and Outreach and Foster Collaboration and National Perspective
- Strengthen Data Management and Sharing
- Diversify Funding Sources and Achieve Sustainable Growth
- Enhance Talent Acquisition and Retention

Discussion

- Looking forward on NCPN – Priorities and areas of focus
- Challenges
  - Funding limitations, close to flat funding for last 6 years
  - Reduced funding for initiatives to preserve operations
  - Focused funding on centers with higher customer demand and industry priority
  - Industry challenges reduces other sources of funding
- Opportunities
  - Technical capacity and expertise
  - Industry support
  - Increase in PPA 7721 funding

Common Themes from Crop Strategic Plans

- Sustainability
- Communication
- Strengthening systems and technology

Management of Diseases Caused by Systemic Pathogens in Temperate and Sub-Tropical Fruit Crops and Woody Ornamentals

WERA-20 Annual Meeting

September 8-10, 2025
(8am to 5pm EST)

Embassy Suites Raleigh/Durham
201 Harrison Oaks Blvd, Cary, NC

AGENDA

Day 1: Monday, September 8th

Field Trip: Pickup at 8am from the hotel

8:30 am	NC State Clean Plant Center: Micropropagation and Repository Unit (MPRU), Raleigh, NC – Dr. Christie Almeyda
9:45 am	NC State Strawberry Production Program, Raleigh, NC – Dr. Mark Hoffmann
11:30 am	Pairwise, RTP, Durham, NC – Dr. Shai Lawit
1:15 pm	Lunch at Boxyard RTP, Durham, NC.
3:00 pm	Union Grove Farm – 7203 Union Grove Church Road, Chapel Hill, NC 27516
5:00 pm	Back to the hotel

Day 2: Tuesday, September 9th

8:00 am Registration

8:30 am Welcome and Opening Remarks
- Dr. Carolyn Young, DEPP Department Head, NC State University

8:40 am Meeting logistics and introductions
- Dr. Christie Almeyda, NC Clean Plant Center Director, NC State University

8:50 am **Business Meeting**
- Dr. Naidu Rayapati, Administrative Advisor, Washington State University Next year's location and host (secretary for this meeting)
- Dr. Amer Fayad, National Program Leader, USDA-NIFA Update

Main Program

9:30 am	Two decades of research on grapevine viruses in Washington State – Naidu Rayapati, Washington State University
9:50 am	A new virus identified in wine grapes – Alex Karasev, University of Idaho
10:10 am	Viral diseases affecting cold-hardy grapes in MN – Joanna Serrano-Salgado, University of Minnesota
10:30 am	Validating multiplex HTS pathogen detection – Kitty Cardwell, Oklahoma State University
10:50 am	Coffee Break
11:05 am	Fruit Tree Report at the USDA APHIS PGQP – Oscar Hurtado-Gonzales, USDA APHIS PPQ PGQP (remote)

11:25 am	Interventions to reduce the entry and spread of pathogens in Fruit Tree Production Systems in Washington State – Segun Akinbade, WA State Department of Agriculture Plant Protection
11:45 am	CFIA Centre for Plant Health: 2024-25 Update - Nomatter Chingandu, Canadian Food Inspection Agency
12:05 pm	Lunch
1:30 pm	Viruses and viroids of peach and blackberry in SC – Elizabeth Cieniewicz, Clemson University (remote)
1:50 pm	Arkansas Update: from clones to clean plants – Ioannis Tzanetakis, University of Arkansas
2:10 pm	California Citrus Report – Georgios Vidalakis, University of California, Riverside
2:30 pm	FPS Report – Maher Al Rwahnih, Foundation Plant Services, University of California, Davis.
2:50 pm	Minnesota Report – WERA 20 – Robert Alvarez-Quinto, University of Minnesota
3:10 pm	Coffee Break
3:25 pm	WERA-20 NC Report – Christie Almeyda, North Carolina State University
3:45 pm	Woody Ornamental Virus trends seen in Agdia's Lab – Deborah Groth-Helms, Agdia Incorporated
4:05 pm	Report on the Diagnostic Assay Validation Network – Kitty Cardwell and Georgios Vidalakis
4:25 pm	Getting to Know Jing Zhou from UH-Manoa – Jing Zhou, University of Hawaii
4:45 pm	Adjourn

Day 3: Wednesday, September 10th

NCPN Cooperators Session

9:00 am Welcome and Orientation to the Day's Session
Dr. Jennifer Nicholson, NCPN Coordinator

Advances in the National Clean Plant Network

9:05 am Progress in the National Clean Plant Network Program - Jennifer Nicholson, USDA-APHIS-PPQ

9:25 am Advances at Foundation Plant Services - Maher Al Rwahnih, University of California, Davis.

9:45 am Advances in Clean Citrus - Georgios Vidalakis, University of California, Riverside

10:05 am Three years in: Building a fully functional Arkansas Center - Ioannis Tzanetakis, University of Arkansas

10:25 am Update from the Oregon Clean Plant Center - Cristian Olaya, Oregon State University

10:45 am Coffee Break

Cross-Network Initiatives

11:00 am NCPN Economics Initiative- Kristen Park, Cornell University

11:20 am NCPN Education/Outreach Initiative - Jennifer Nicholson, USDA-APHIS-PPQ

11:40 am NCPN Quality Initiative - Irene Lavagi-Craddock (remote) and Fatima Osman, University of California, Riverside and Davis

12:00 pm NCPN ADAPT Core - Karl Krist (remote), University of California, Davis

12:20 pm Lunch

1:30 pm Group Discussion - Cross-network needs and areas of collaboration

2:30 pm Coffee Break

2:45 pm **Future Strategies**

- Overview of NCPN Strategic Planning efforts, challenges and opportunities - Jennifer Nicholson
- Group Discussion - Strategies for advancing the NCPN and sustaining into the future

4:00 pm Recap and Wrap-up

4:30 pm Adjourn