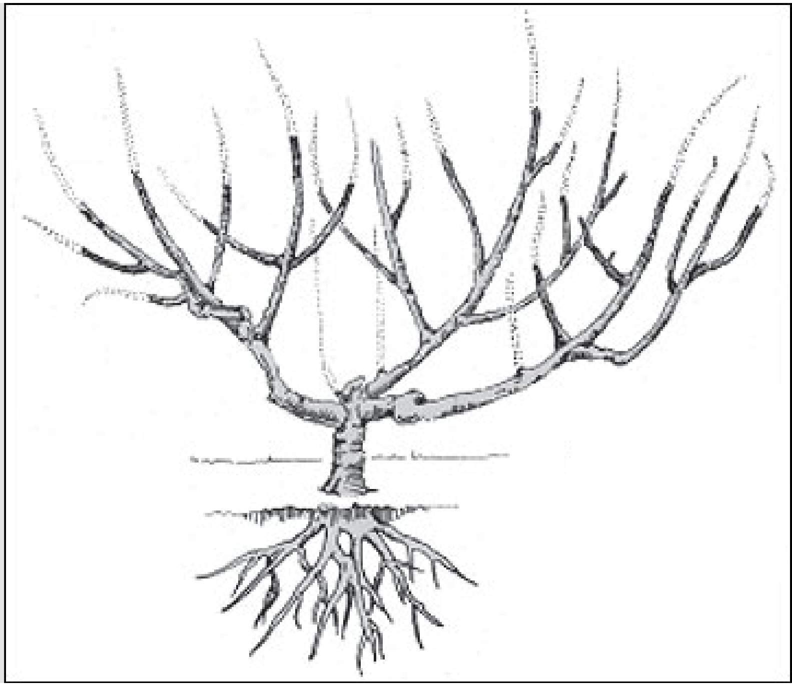
Getting to the Root of ‘Flordaguard’ Rootstock Resistance:

Rootstock Alternatives and Current Efforts



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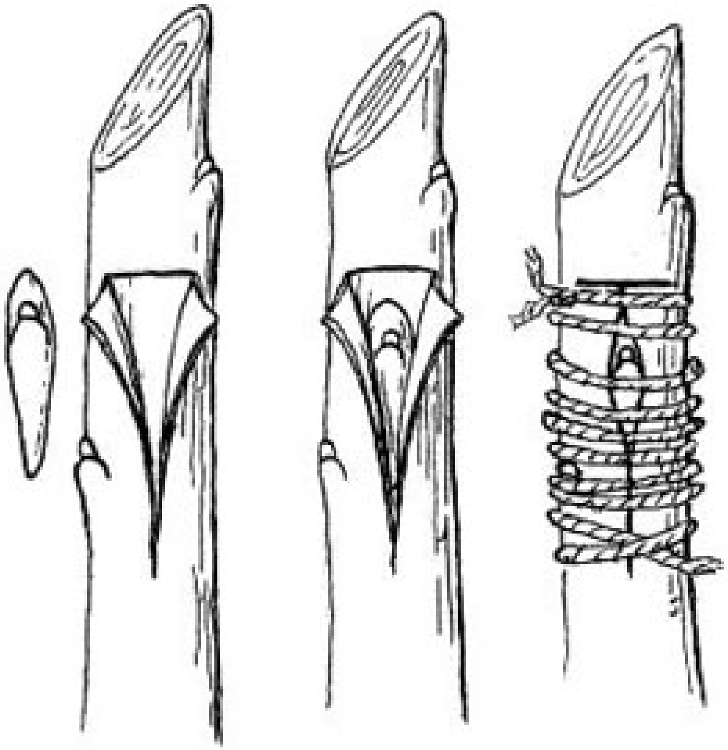
Horticultural Sciences Department

UF Stonefruit Field Day
April 22, 2014
PSREU, Citra

UF UNIVERSITY of FLORIDA

Peach trees are composite genetic systems

- Genetically distinct scion
  - Unique fruit quality
  - Low-chill adaptation
- Genetically distinct rootstock
  - Locally-adapted
  - Nematode-resistant



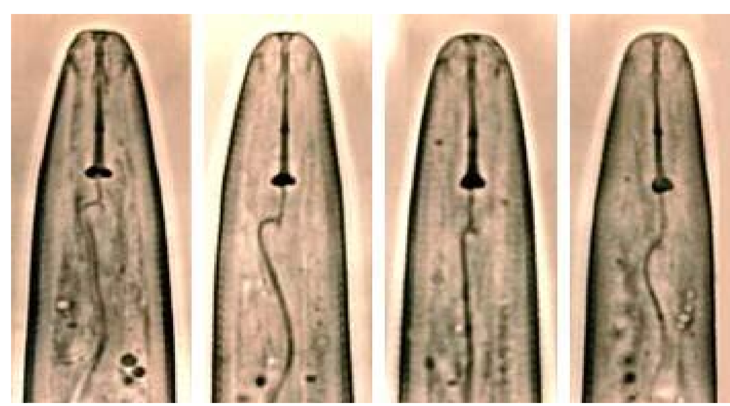
Root-knot nematodes (RKN), *Meloidogyne* spp., are parasitic to many agricultural crops

- **Four most common RKN species:**

1. ***M. incognita*** **(MI) – the Southern RKN**
2. ***M. javanica*** **(MJ) – the Javanese RKN**
3. ***M. arenaria*** **(MA) – the peanut RKN**
4. ***M. hapla*** **(MH) – the Northern RKN**

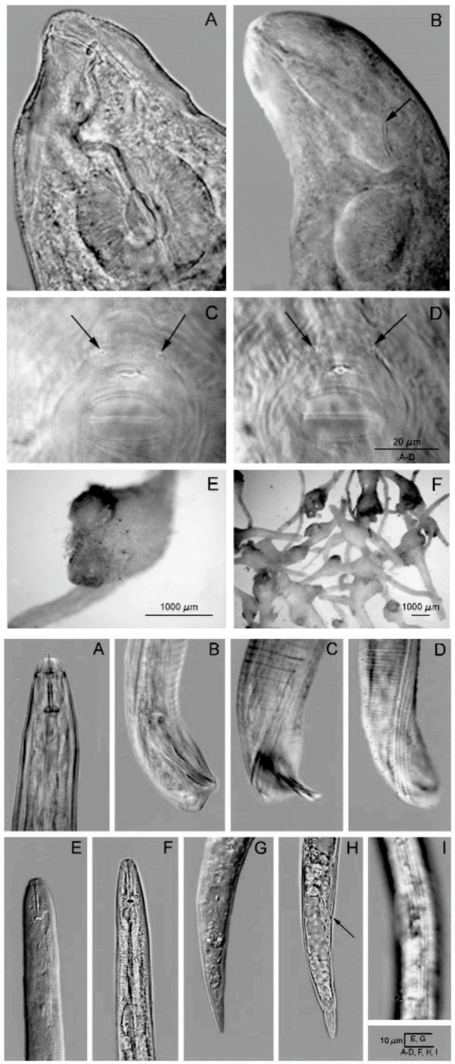
Three major species present in most areas with tropical and Mediterranean climates (species 1–3)

Light micrographs of the anterior end of second-stage juveniles



http://nematode.net/NN3_frontpage.cgi?navbar_selection=speciestable&subnav_selection=Meloidogyne_javanica

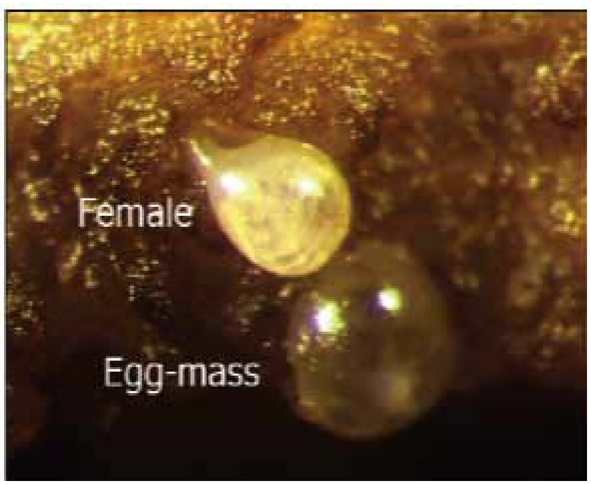
Problem: the peach root-knot nematode infects known resistant peach rootstocks



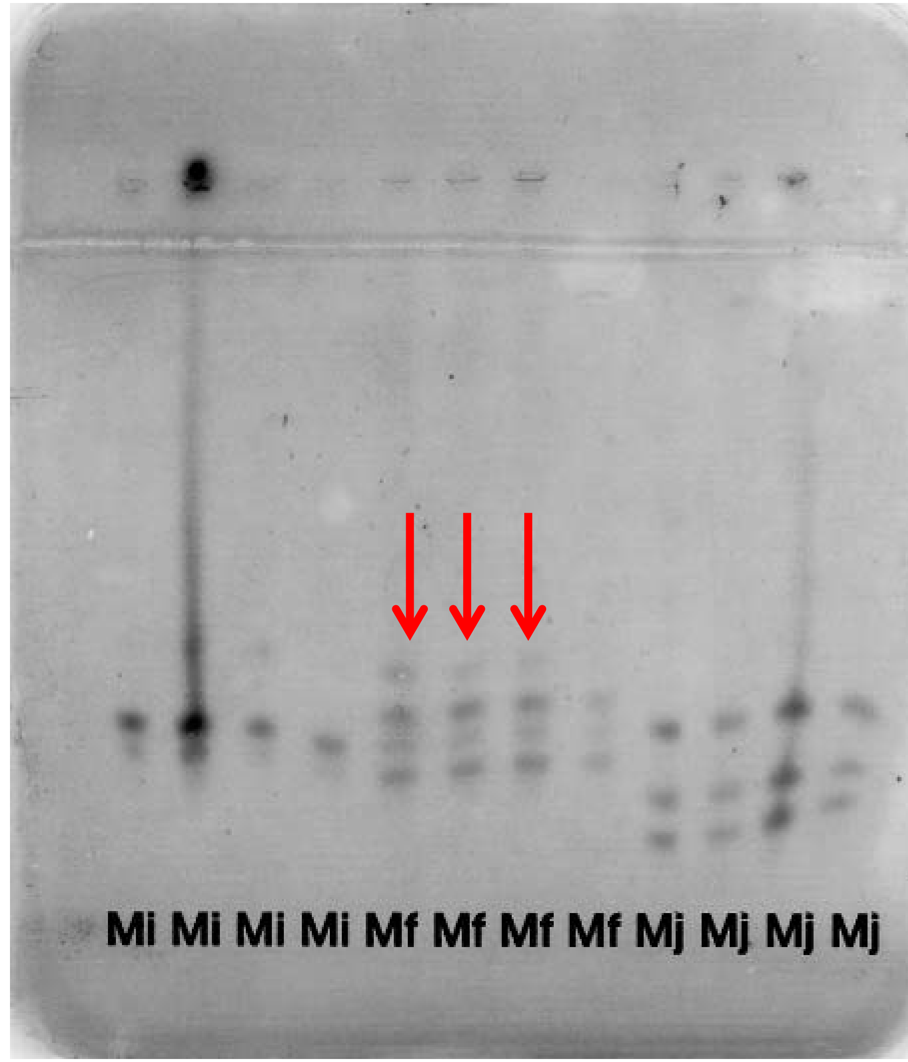
- *Meloidogyne floridensis (MF*)
  common name: peach root-knot nematode
- Occurs only in Florida - first detected by Dr. Ralph Sharpe in 1960s in Gainesville, FL
- Initially identified as *M. incognita* Race 3
- Characterised as a new species based on morphology and unique esterase isozyme pattern (Handoo et al., 2004; Carneiro et al., 2000)
- Wide host range and overcomes nematode resistance of 'Nemaguard', 'Okinawa', 'Guardian' and 'Nemared' rootstocks

Comparison of esterase dehydrogenase profiles of *Meloidogyne floridensis* n. sp. (Mf) with that of *M. incognita* (Mi) and *M. javanica* (Mj).

Adult female (J4) and egg mass



http://pubs.ext.vt.edu/444/444-107/444-107.html

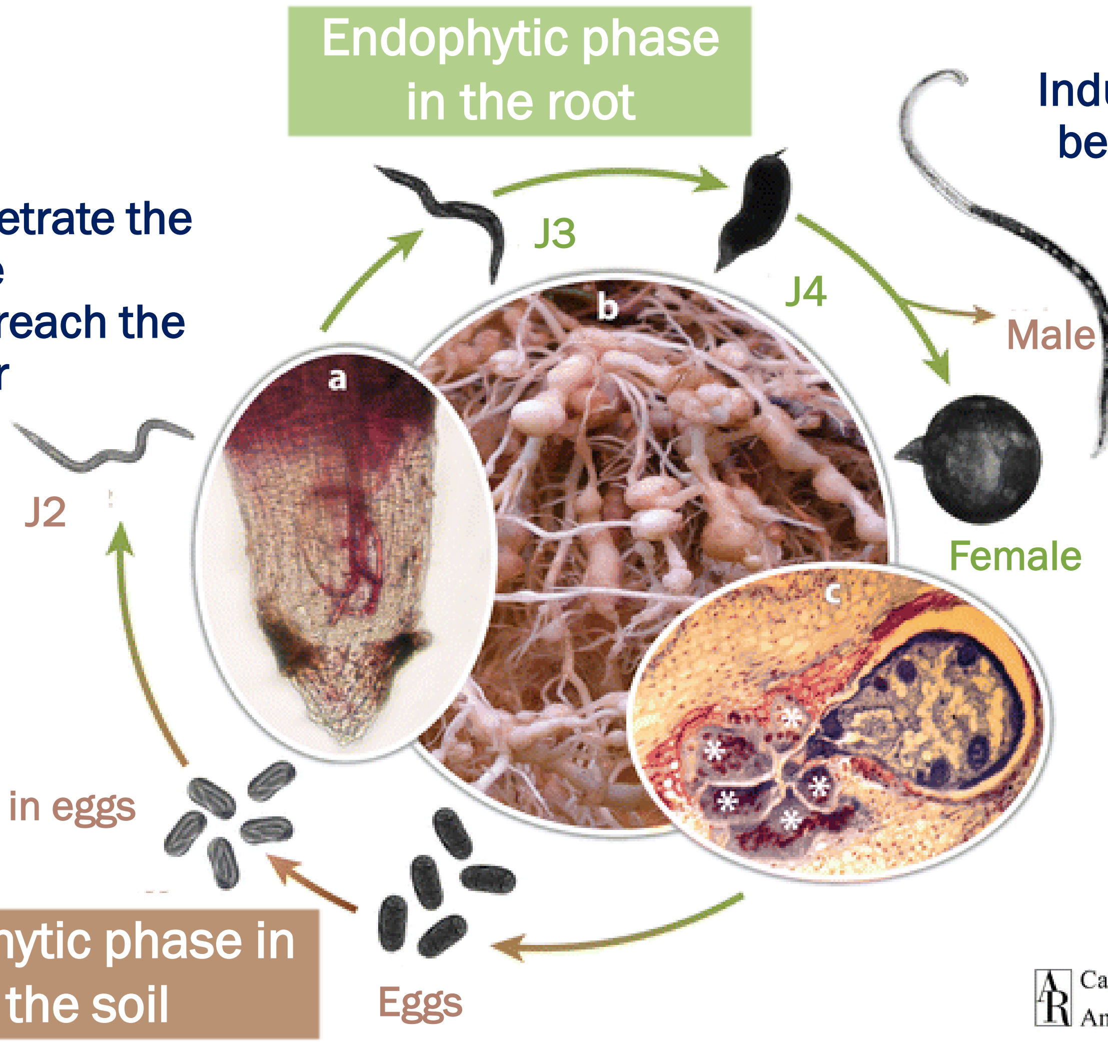


Source: Carneiro, R. M. D. G., M. R. A. Almeida, and P. Queneherve. 2000. Enzyme phenotypes of *Meloidogyne* spp. populations. Nematology 2:645–654.

Root-knot nematode infestation life-cycle

I. Invasion
J2 juveniles penetrate the root and migrate intercellulary to reach the vascular cylinder

II. Induction
Induces feeding cells, becomes sedentary



III. Nutrient Acquisition
Vascularization of feeding site, nematode matures to adult

Castagnone-Sereno P, et al. 2013. Annu. Rev. Phytopathol. 51:203–20

Damage Caused by RKN in Peach

Tree stunting and reduced vigor

Damage Caused by RKN in Peach

- Early defoliation and reduced foliage resulting in the production of unpalatable "stress fruits"

Damage Caused by RKN in Peach

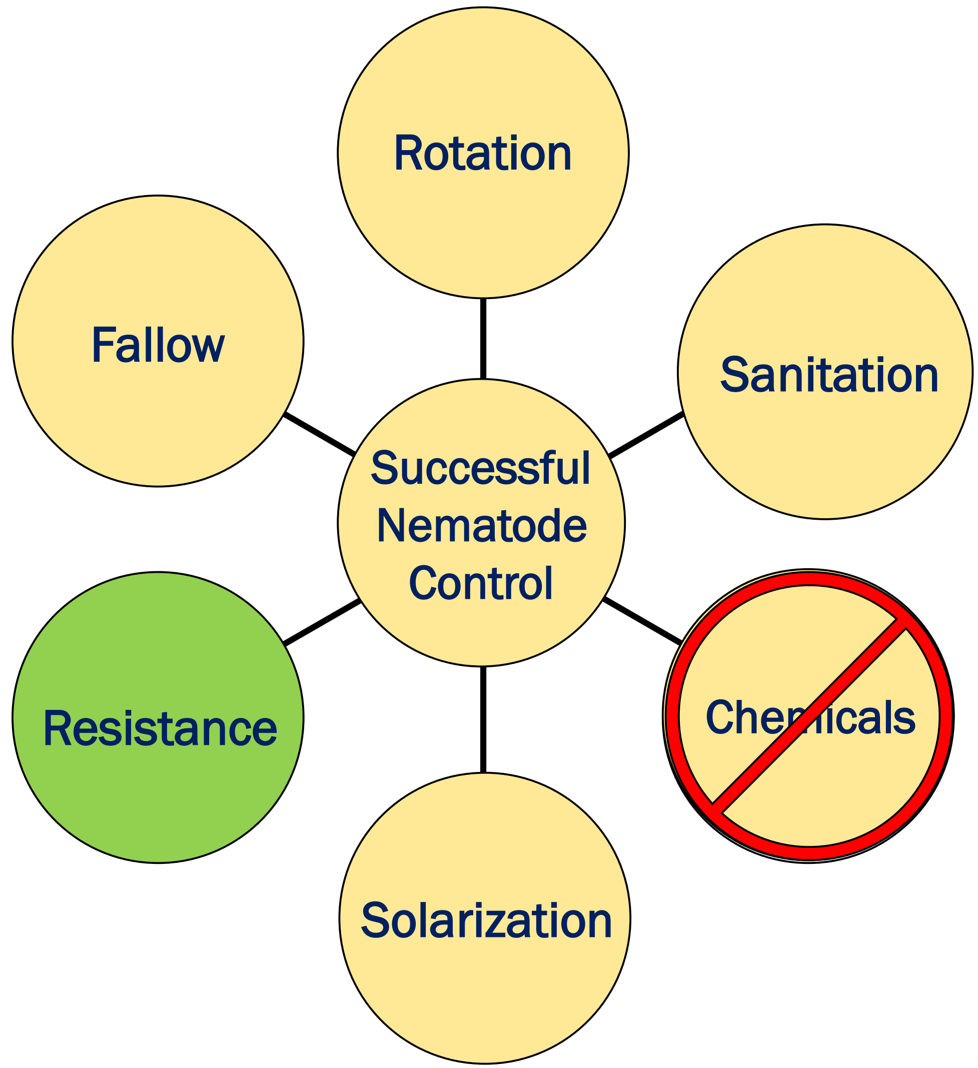
Galled roots

http://www.freshfromflorida.com/pi/enpp/triology/5001/triology_5001_nematology.html

Nematode Management

No Quick Solutions…

- Can't eliminate nematodes
- No post-plant nematicides available for many crop-nematode combinations
- No single practice will control nematodes, so two or more control methods must be used
- RKN-resistant cultivars – effective against nematode genotypes



- Three commonly used rootstock Nemaguard, Guardian, Nemared, are susceptible to *Meloidogyne floridensis*

- Nemaguard and Nemared require more winter chill for proper fruiting

- **Flordaguard has improved root-knot nematode resistance and low-chill adaptation**
  - Used as standard rootstock for low-chill peach production in root-knot nematode infested non-alkaline soils

http://hos.ufl.edu/extension/stonefruit/stone-fruit-varieties

How do we manage the disease while maintaining production and staying competitive?

Current Efforts

I. Molecular characterization of resistance in Flordaguard

- Understand the genetic nature of resistance to *MF* in 'Flordaguard' peach
- Identify SSR markers associated with resistance against *MF* in various segregating populations

II. Evaluation of horticultural performance of peach rootstocks

- Identify potential rootstocks with resistance to *MF*

Resistance spectrum of main *Prunus* RKN-resistant sources

(Claverie et al., 2004; Van Ghelder et al., 2010; Cao et al., 2011)

Rootstock	MA	MI	MJ	MF	RKN resistance genotype
Almond *(P. dulcis)*					
'Alnem1'	R	S	R	S	R_{Mja} / R_{Mja}
Peach (*P. persica*)					
'Shalil'	R	R	S	S	R_{Mia} / r_{Mia}
'Nemaguard', 'Nemared'	R	R	R/S*	S	R_{MiaNem} / R_{MiaNem}
$(G \times N)_{15}$ = 'Felinem"	R	R	R/S*	S	R_{MiaNem} / r_{MiaNem}
Myrobalan plum (*P. cerasifera)*					
Accession P.2175	R	R	R	R	*Ma* / *ma*
Japanese plum (*P. salicina*)					
Accession J.222	R	R	R	R	R_{jap} / r_{jap}
Wild peach (*P. kansuensis*)					
'Honggengansutao'	--	I**	--	--	*PkMi* *tested only on MI; possibly an allele of* R_{Mia}

*R/S – variable behaviour in function of *M. javanica* isolates

**I = Immune

Parental Genotypes for the Crosses

300 cu

Flordaguard
(*P. persica*)
rr

and hybrids
with *P. kansuensis*
Rr

Wild peach
(*P. kansuensis*)
rr

'Okinawa'
(*P. persica*)
RR

and hybrids
with *P. kansuensis*
Rr

325 cu

'UF Sharp'
(*P. persica*)
RR

Generating the mapping populations for phenotyping

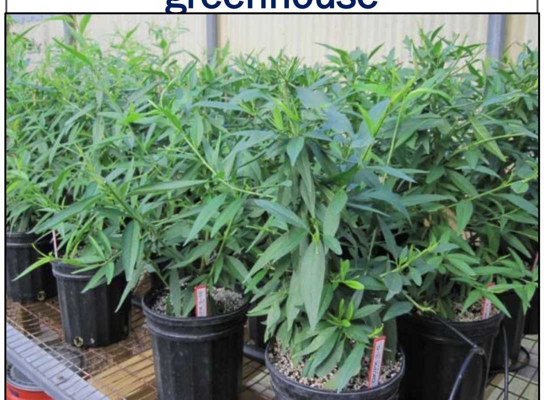
1
Pollen collection
(late Jan – early Feb)

2
Manual pollinations
(late Jan – early March)

3
Fruit harvesting & seed extraction
(April – May)

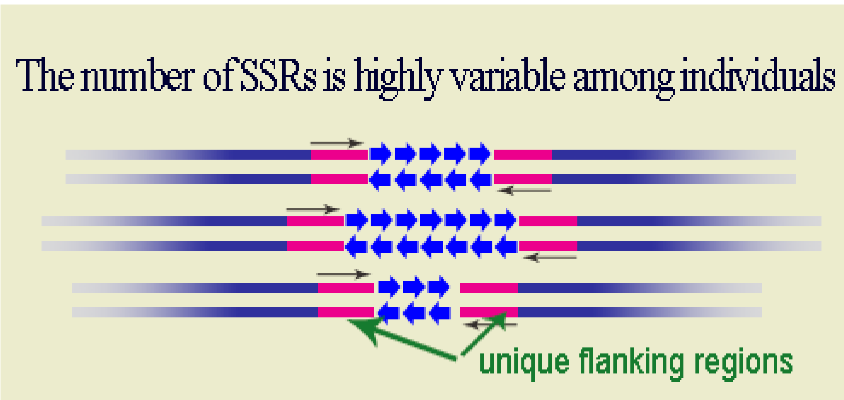
4
Seed stratification and germination
(Jun – Aug)

5
Growing plants in greenhouse

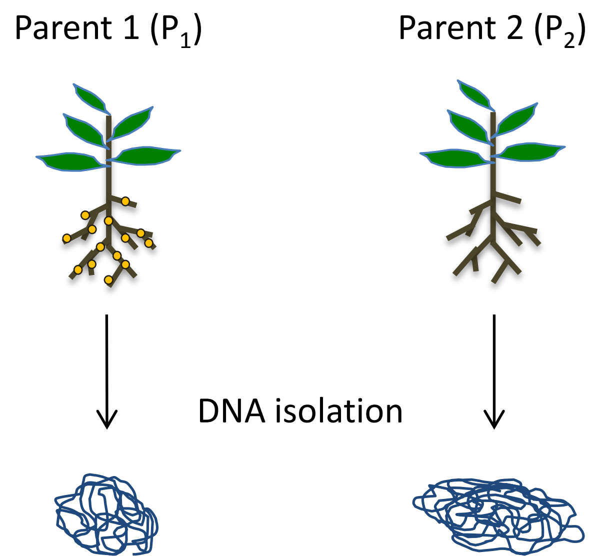


6
Screening for MF resistance
(Dec - Mar)

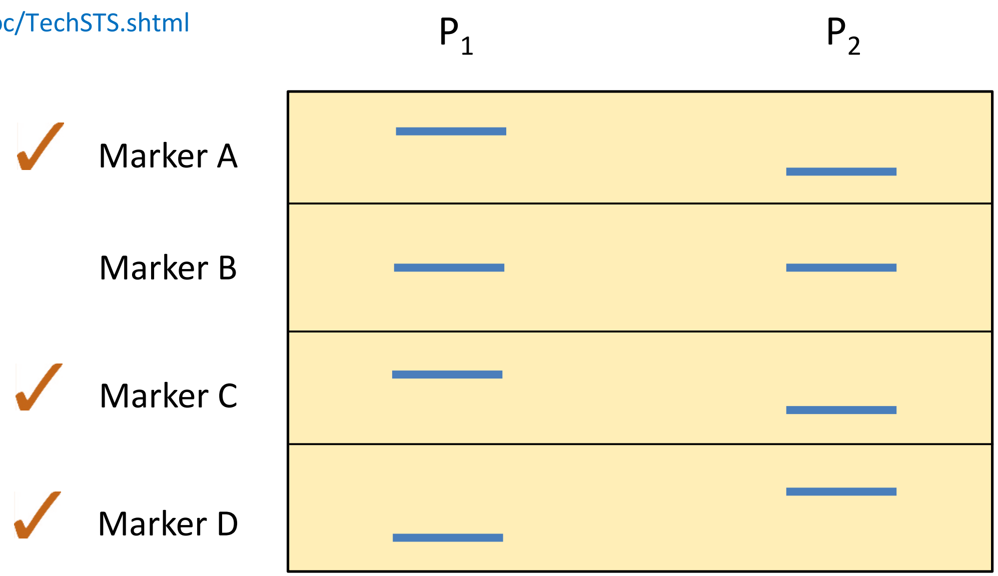
Screening of parental genotypes with SSR markers



https://www.ncbi.nlm.nih.gov/projects/genome/probe/doc/TechSTS.shtml



Screening parental genotypes with molecular markers



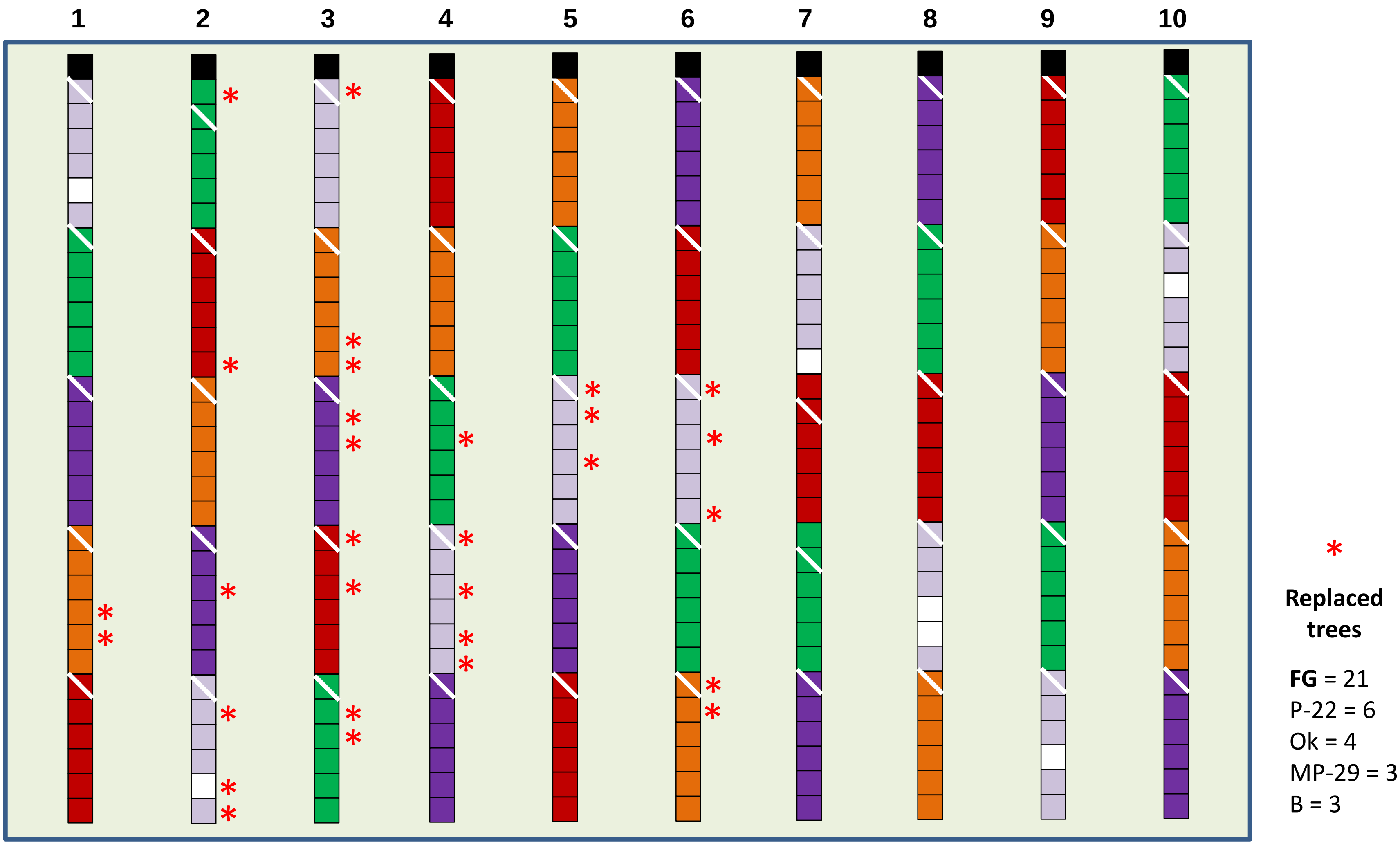
Markers A, C, and D can differentiate the two parental genotypes

Current Efforts…

II. Evaluation of horticultural performance of peach rootstocks

- Identify potential rootstocks with resistance to *MF*

Peach rootstock field evaluation for Resistance to *M. floridensis*



Peach Rootstocks

Rutgers Tomato **Scion:** UFSun Flordaguard Barton Okinawa MP-29 P-22

Experiment Design

- Location: Citra, Florida
- Plot size: 200 ft x 130 ft
- Design: RCBD with 5 subsamples per treatment
- No. of rows (replicates): 10
- Spacing: 4ft between trees, 20ft between rows
- Treatments: Flordaguard (*R*), Barton (*R*), Okinawa (*S*), and 2 new USDA rootstocks (*R*)

Plants established in microplots

Photo courtesy of Dr. Andrew Nyczepir, USDA-ARS Byron, Georgia

Setting up the microplots

Auger was used to excavate holes

Trenches at plot borders for installation of sprinkler irrigation system

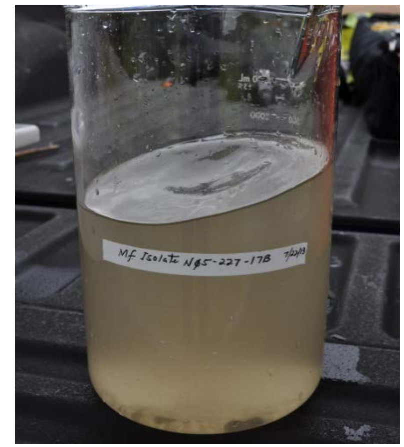
Microplots were laid out in rows and fumigated soil was backfilled into pots

Budding on ‘Barton’ and ‘MP-29’

MP-29

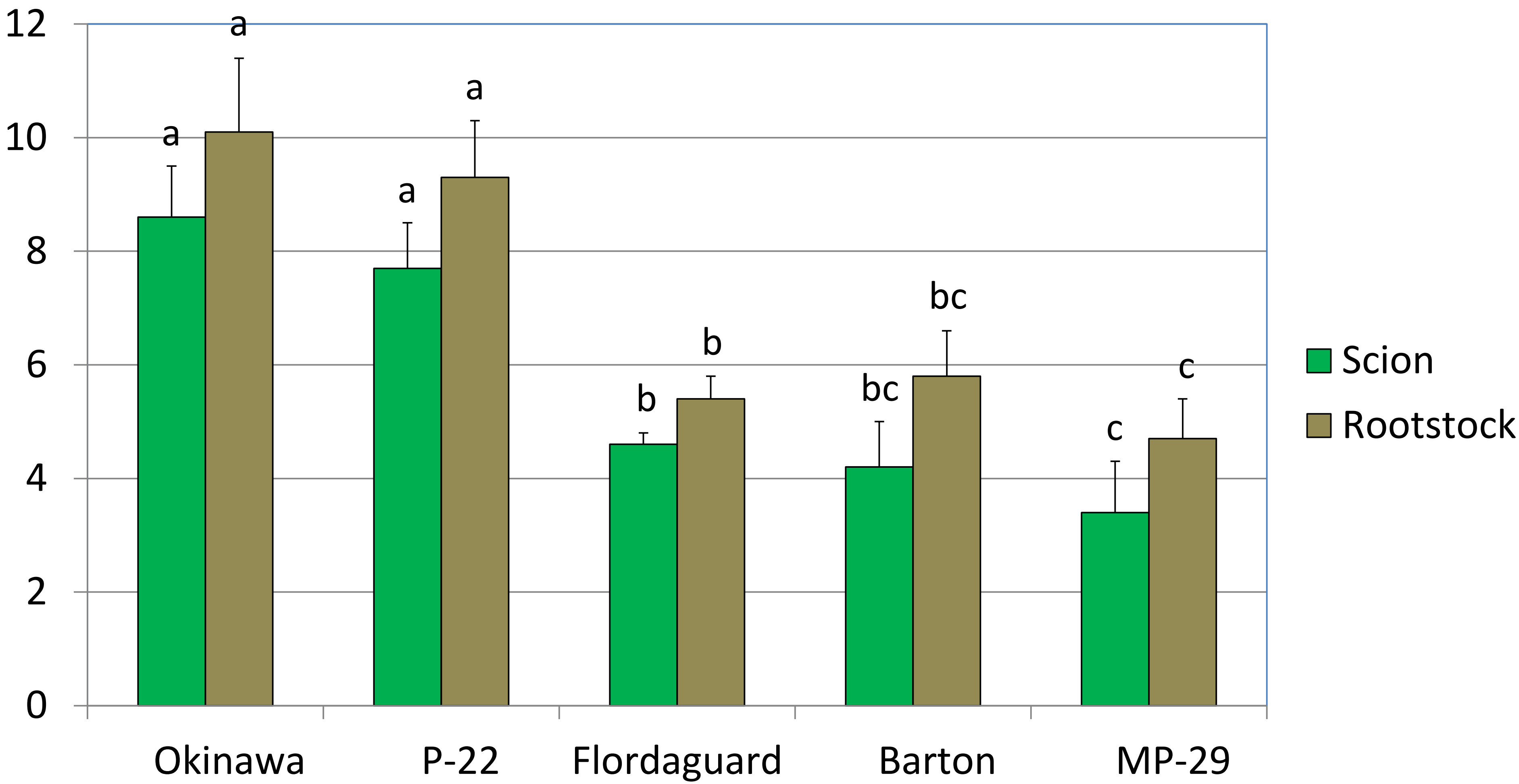
Barton

Nematode inoculation for resistance evaluation

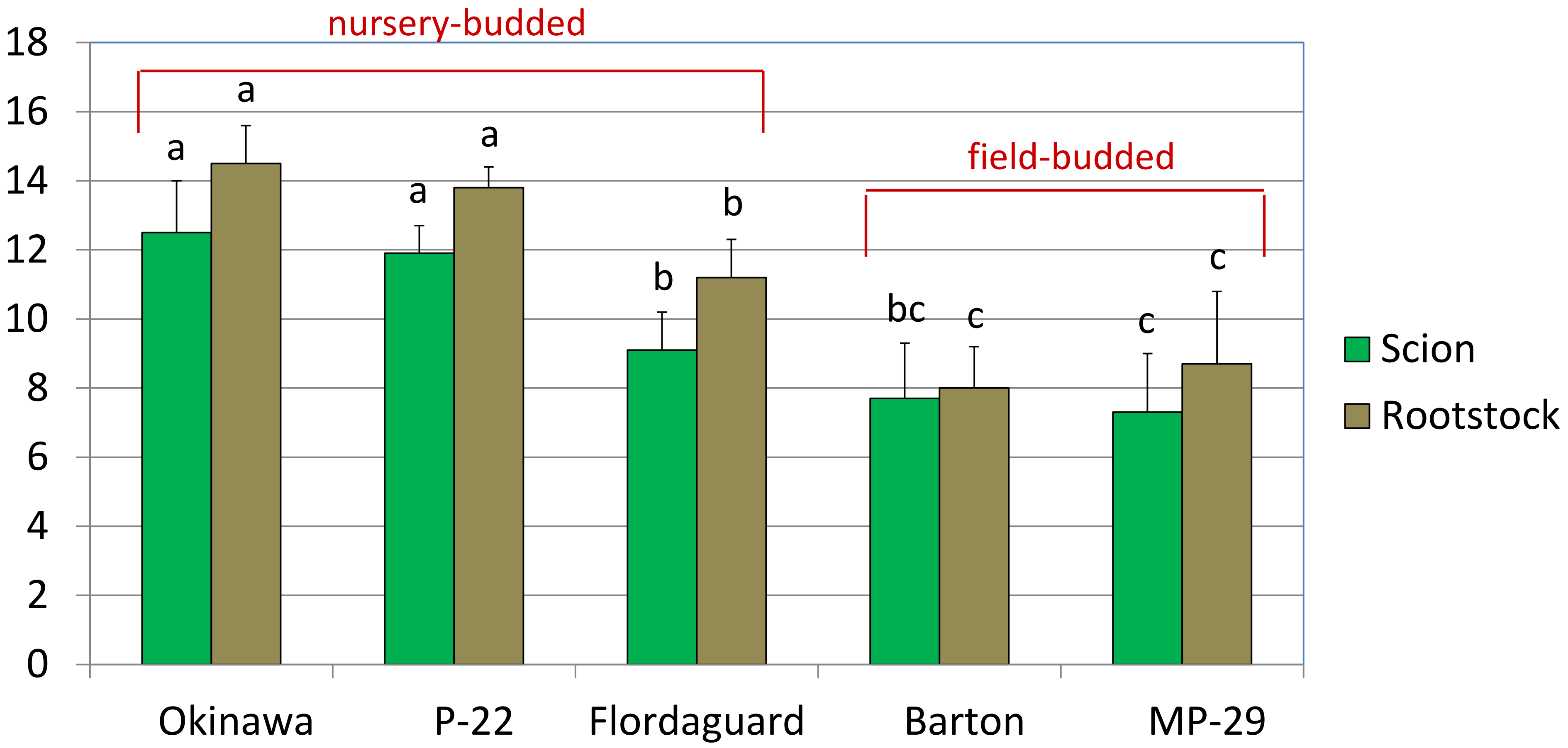


Total of 45,000 juvenile nematodes inoculated per plant

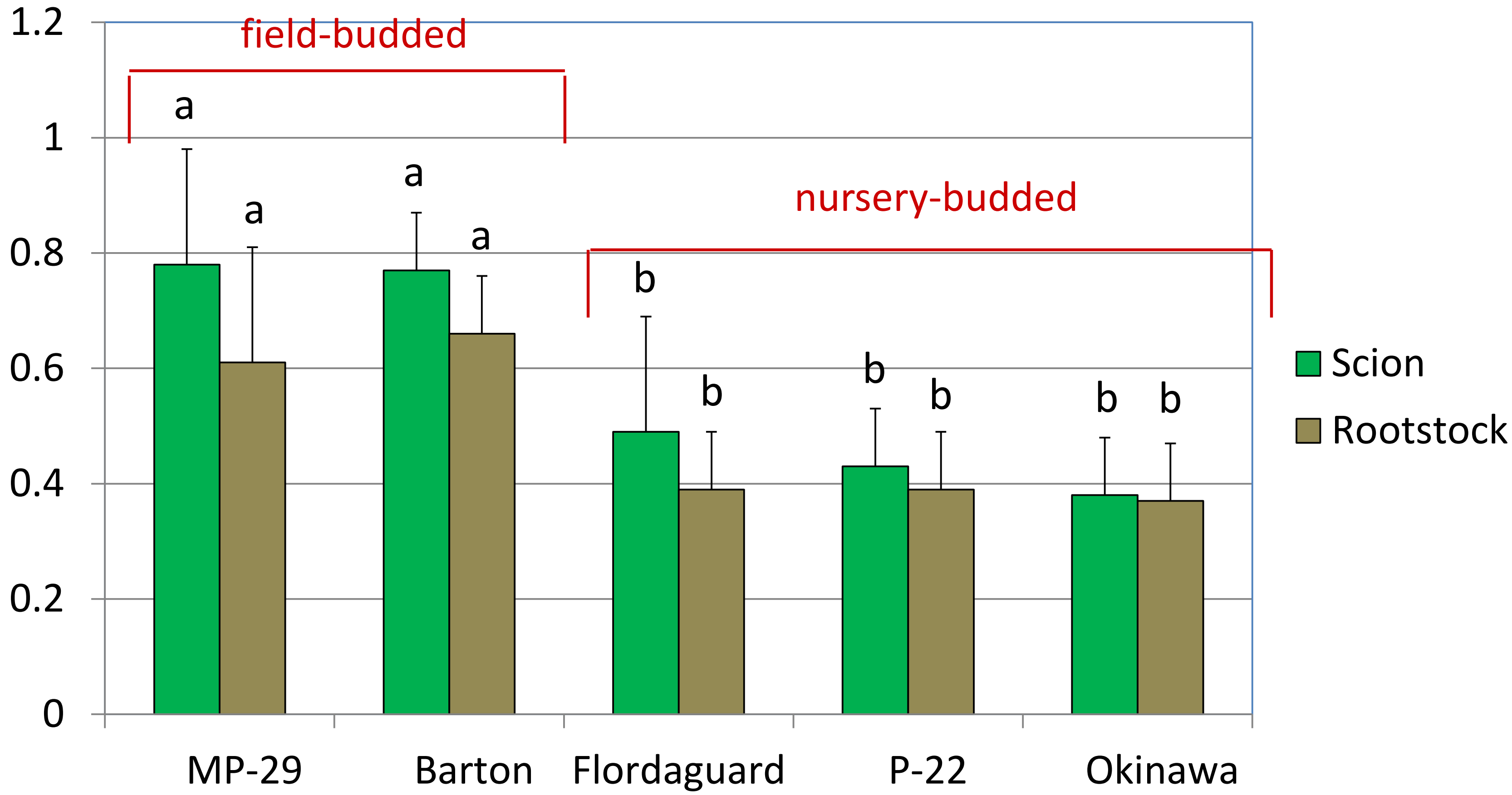
2013 Scion and rootstock trunk circumferences (2 inches above and below graft union) of grafted trees in field microplots at Citra, FL.



Scion and rootstock trunk circumferences (2 inches above and below graft union) of grafted trees in field microplots



Relative growth rates of scion based on trunk circumference (2 inches above graft union) at the end of 2013 growing season in field microplots



Pearson correlations between the scion trunk circumference and selected growth (n = 50)

*, P=0.02; **, P=0.001.

Parameter		Correlation coefficient, r
Scion trunk circumference	Pruning weight	0.83**
	Tree height	-0.28**
	Tree spread	0.88**
	Scion trunk relative growth rate	-0.33*

More studies underway…

II. Testing of 'Flordaguard' for durability of resistance against four main root-knot nematodes – *M. arenaria, M. incognita, M. javanica, and M. floridensis*

III. Histological characterization of resistance in 'Flordaguard'

Histological characterization of resistance

Terminal root fragments, 10 mm long

1, 2, 3, 5, 8, 12, 17, 23, 30, 40 d post-inoculation

Examine for nematode penetration and root galling

Chemical fixation with 2% glutaraldehyde

overnight

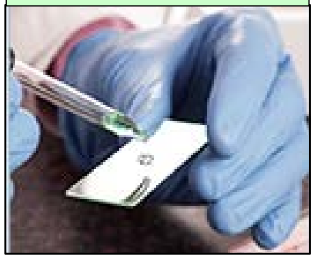
Processing – dehydration through graded ethanol series and clearing

2 d

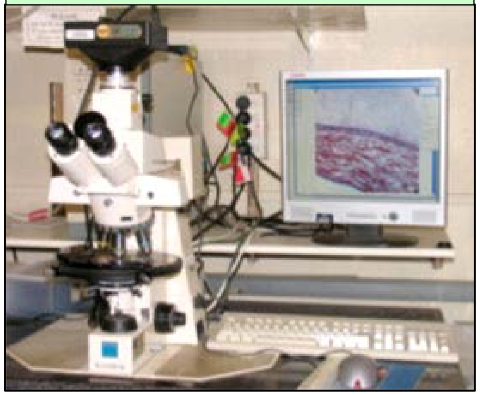
Infiltration and embedding with epoxy resin

Sectioning, 5 *um*

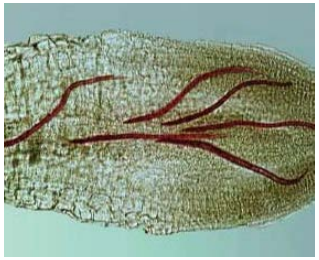
Staining



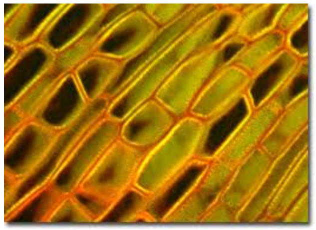
Microscope examinations



Stained slides



Unstained slides for phenolic compound examination

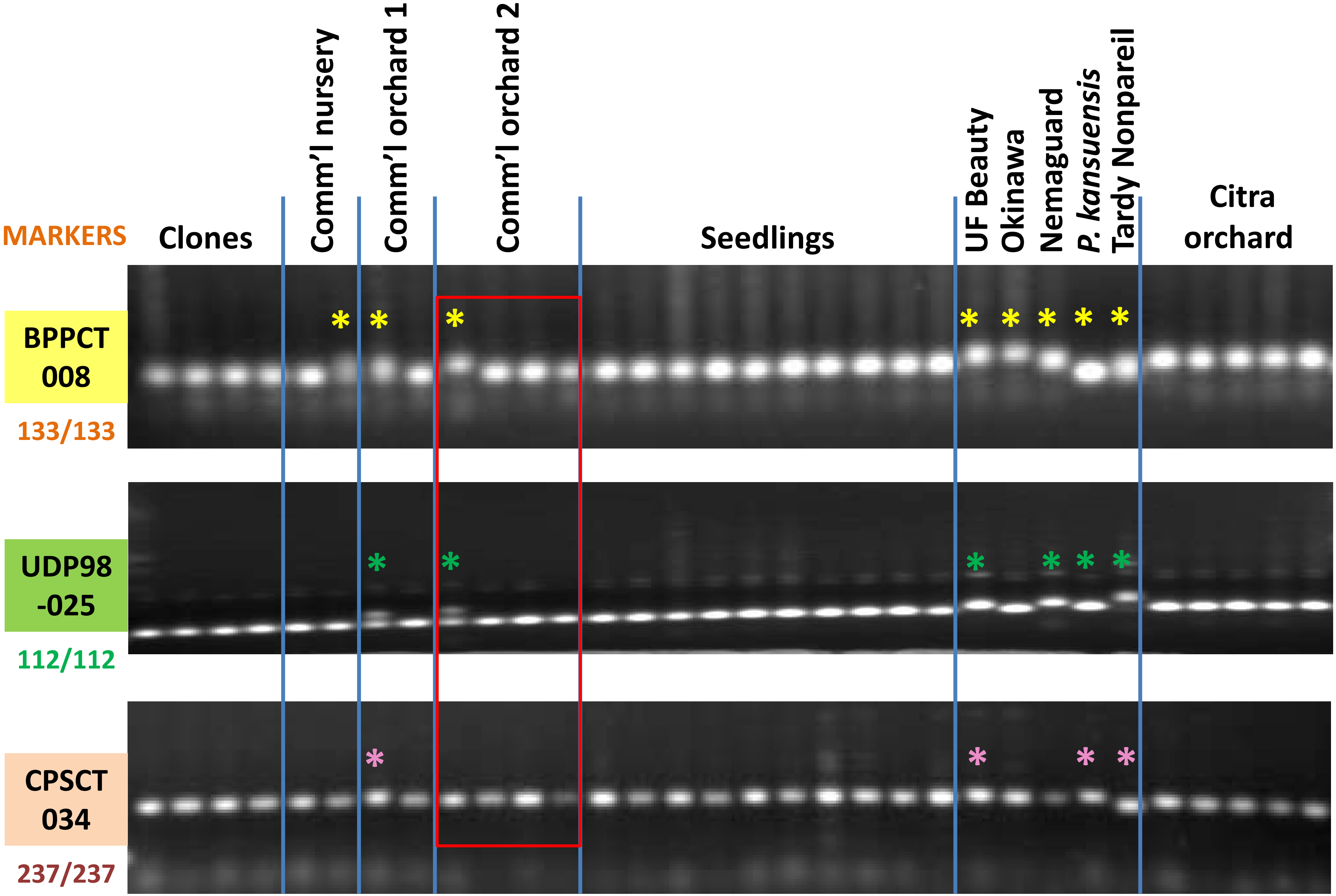


Nematode management requires a concerted effort…

- **UF Stonefruit breeding program**
  - Continued efforts to develop improved rootstocks for the peach industry

- **Nurserymen**
  - Maintain nematode-free nursery stock
  - Ascertain rootstocks' trueness-of-type

- **Growers**
  - Monitor nematode numbers
  - Report any signs of nematode infestation
  - Send soil/root samples to nematode diagnostics lab to identify nematode species


MARKERS
Clones
Comm'l nursery
Comm'l orchard 1
Comm'l orchard 2
Seedlings
UF Beauty
Okinawa
Nemaguard
P. kansuensis
Tardy Nonpareil
Citra orchard
BPPCT 008
133/133
UDP98 -025
112/112
CPSCT 034
237/237

Assessing trueness-of-type

OUTCROSS!

TRUE-TO-TYPE

(red color fades when exposed to warm temperatures)

Resistance-breaking nematodes?

Collecting root and soil samples for nematode diagnostics

Send samples to: UF Nematode Assay Lab

nematology.ifas.ufl.edu

Acknowledgment

Florida Department of Agriculture and Consumer Services (FDACS) Specialty Crop Block Grant

Lineage of ‘Flordaguard’ Rootstock

Shau Thai op Chico 11
(PI 65821)

x op H-91 op Fla. 12-9 op Fla. 17-64
outcross

Prunus davidiana
(C-26712)

Fla. 4-115 op Fla. 14-11
(Flordaguard)

either
(N.J. 5106137 x Okinawa)op
or
(Okinawa x Rancho 23/32)op

op = open pollination
x = unnumbered selection
N.J. 5106137 = J. H. Hale x (Elberta x Rutgers Redleaf)