



Insecticide mode of action rotation for psyllid control in Florida

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IPM- tactics used to manage ACP

Research Areas

1. Biological control
2. Chemical control
3. Other: Repellents, Attractants, Barriers
4. Technologies in progress: NuPsyllid etc

Bottom line: Integrated pest management as employed to date has not suppressed psyllids sufficiently low to prevent disease spread.

2. Chemical control

- a. Conventional insecticide efficacy
- b. Organic treatments
- c. Timing of treatments for adult overwintering and flush
- d. Border treatments
- e. Coordinated treatments over large areas
- f. Systemics for nymph control
- g. Anti-feedant characteristics are essential
- h. Secondary pest outbreaks
- i. Resistance management

1. Chemical Control (continued):

- Uptake of the bacterium by nymphs is more efficient than adults, because they are often located near where the bacterium was deposited and feed at that site for long periods of time. **Systemic insecticides** are especially important for controlling nymphs, but not effective in all situations (soil type, irrigation method, tree size, *insecticide resistance*).
- Young tree **replants**, especially in Florida where the bacterium is everywhere, need a high level of protection if they are going to become fruit bearing
- Insecticides with **anti-feedant** qualities are essential

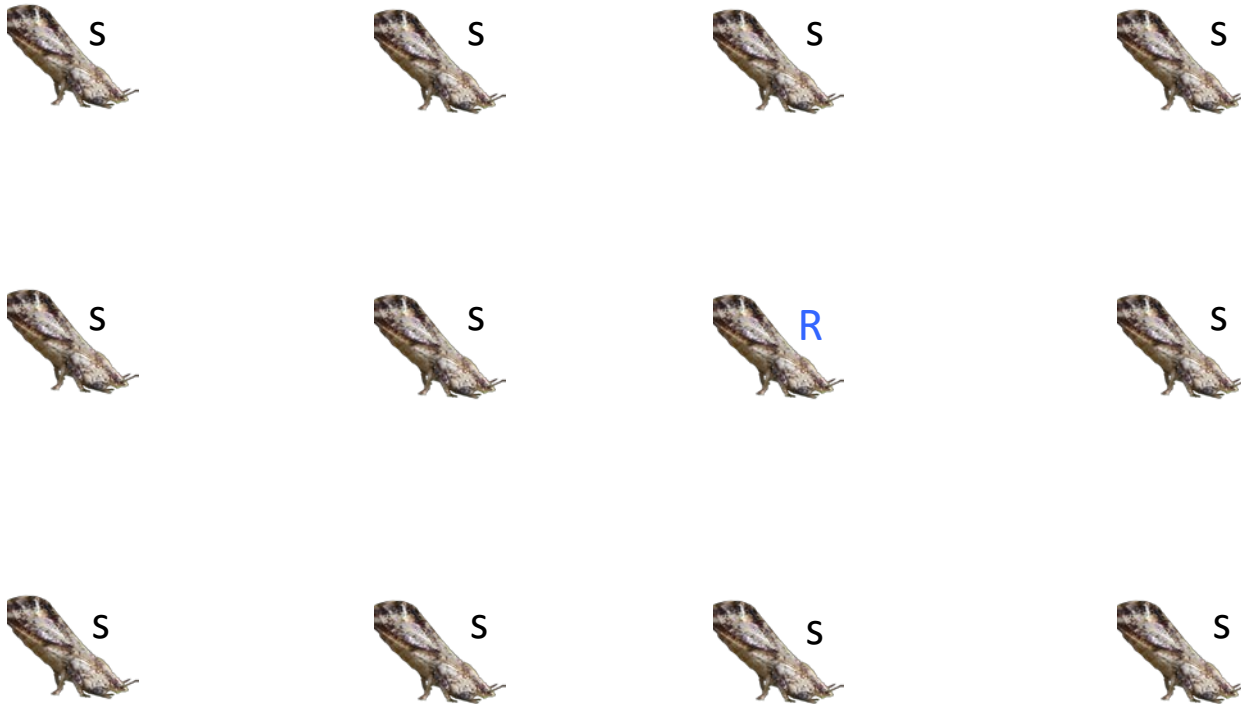
| Tree size | Jan | Feb | Mar | Apr | May | Jun | Jul | Aug | Sep | Oct | Nov | Dec |
|-----------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Reset (<3') | P | | A | A | | B | B | | A | A | | P |
| 1-2 yr (3-5') | P | | A | B | | B | B | | B | A | | P |
| 3-5 yr (5-9') bearing | | | | P | | | | A | | | | |

A= Admire (imidacloprid); B=Belay (clothianidin); P=Platinum (thiamethoxam); Products are positioned for use at certain times of the year based on water solubility and likelihood for significant rain events.

Types of resistance possible in Florida populations of ACP

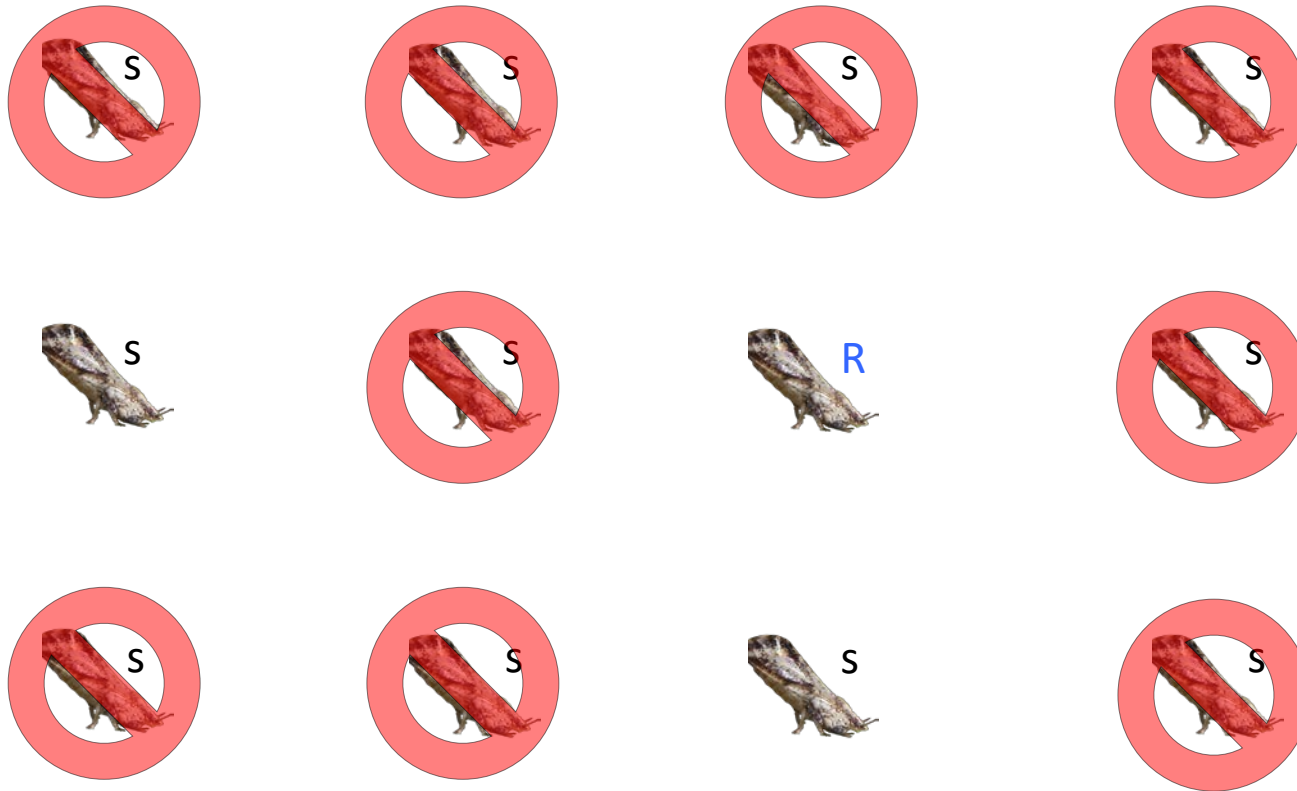
- Multiple resistance: resistance to multiple insecticides of different classes by multiple mechanisms.
Consequence of sequential application of insecticides (Pesticide treadmill)
- Cross resistance: resistance to one insecticide leads to resistance to another yet unused insecticide. Usually the two insecticides belong to the same class and share identical or similar mode of action
- Negative cross-resistance: resistance to insecticide A leads to susceptibility to B and vice visa

Evolution of Insecticide resistance How does it occur?



Individuals in a population are never equally susceptible to an Insecticide. Although initially rare, insecticide-resistant **R genotype** is present. Frequency is $1/12 = 0.083$

Evolution of Insecticide resistance. How does it occur?



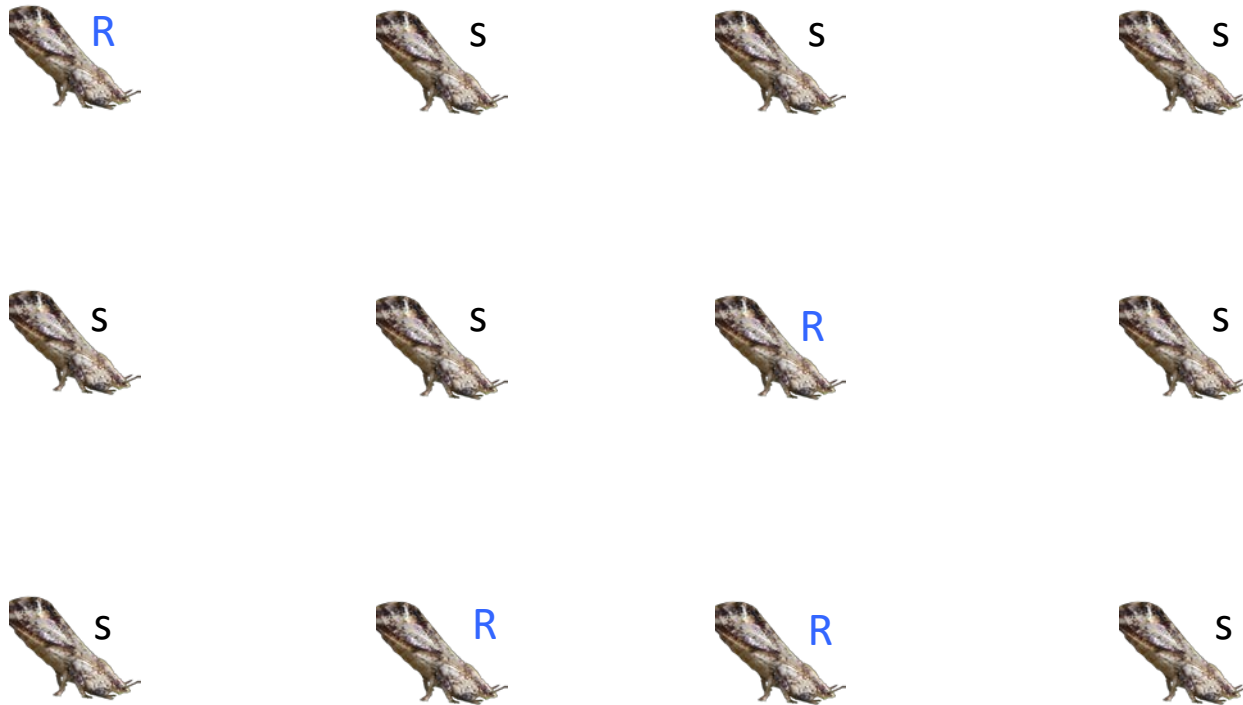
An insecticide is used leaving some individuals insecticide-resistant individuals (**R genotype**) and some susceptible individuals (s genotype)

Evolution of Insecticide resistance. How does it occur?



Now, the frequency of insecticide-resistant **R** genotype is $1/3 = 0.333$

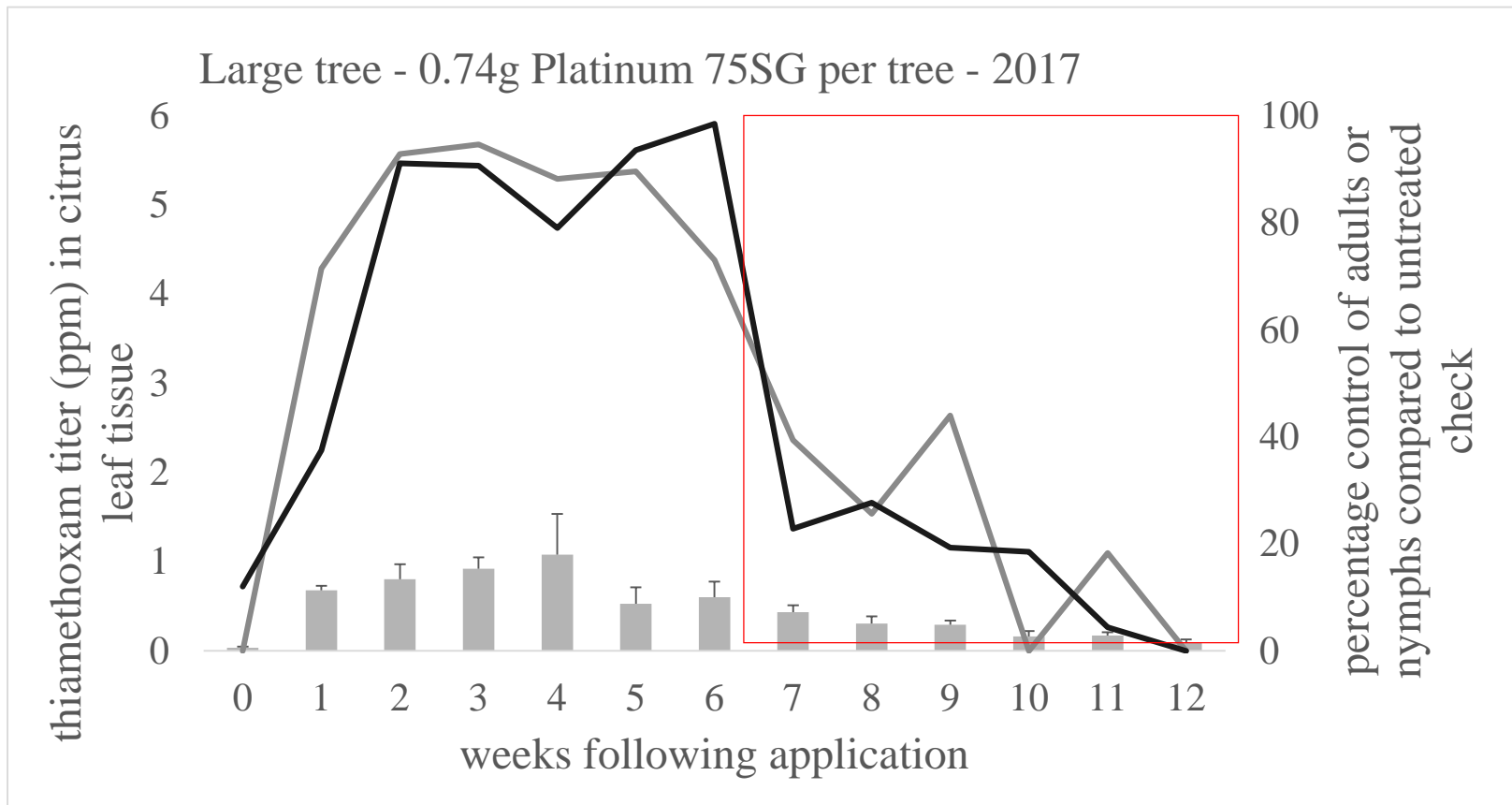
Evolution of Insecticide resistance. How does it occur?



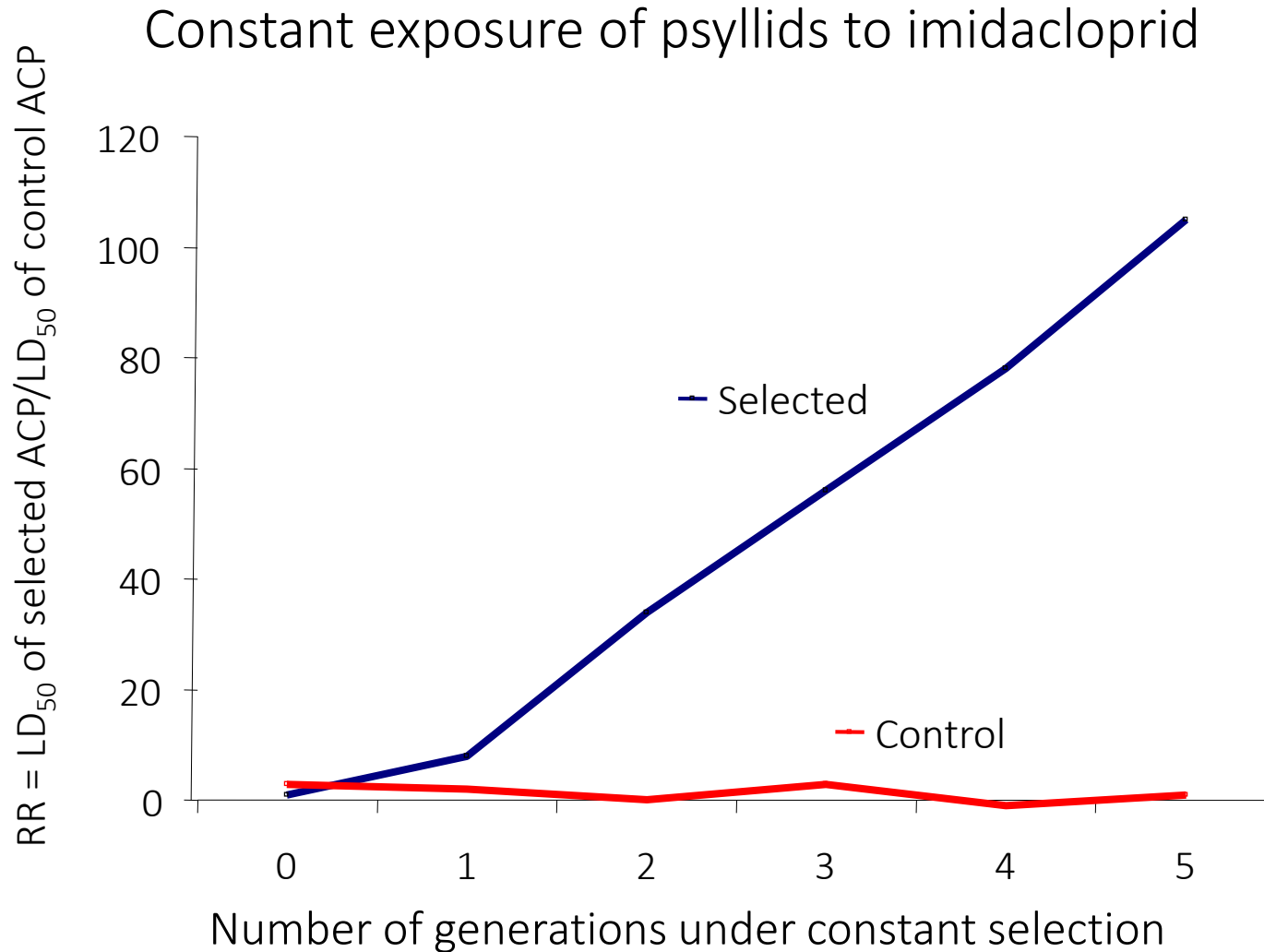
If the **R genotype** reproduces as well as the s genotype, in the next generation the frequency of the **R genotype** will be the same as the survivors in the preceding generation, i.e. **0.333**

Sub-lethal concentrations of neonicotinoids persist long after treatments are effective against ACP—causes selection pressure

18 month old trees



How does it look with ACP under constant selection?



Evolution of insecticide resistance:

- When LD50 ratio ≥ 10 , resistance occurs

- Resistance: occur through insecticide selection

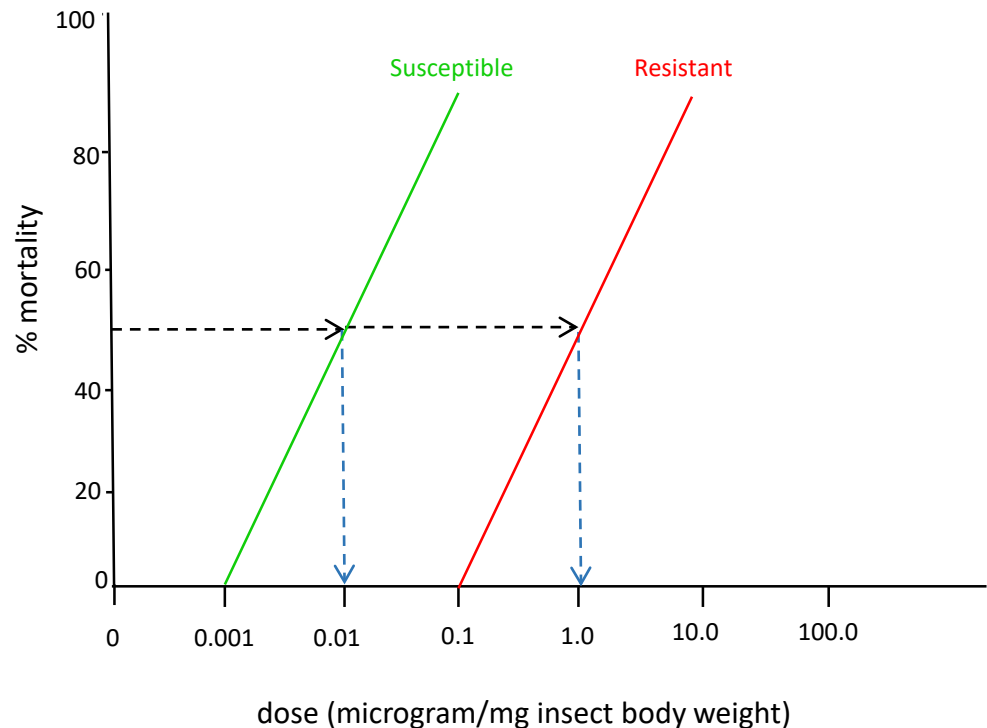
- Selection acts on genetically based variation in susceptibility which arise from:

- Mutation, the source of all new genetic variation

- Genetic recombination that rearranges genetic variation

- Gene flow from populations having different allelic frequencies

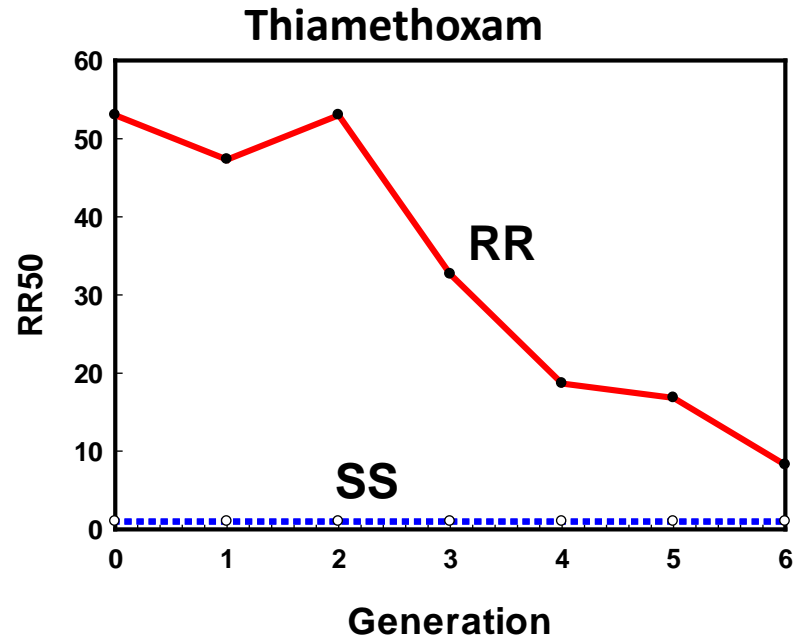
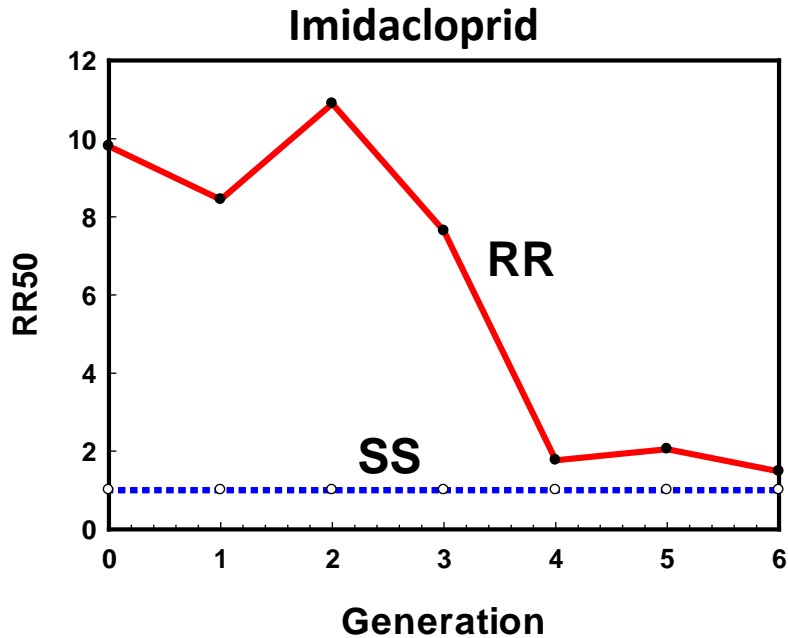
LD₅₀ = A measure of insecticide toxicity



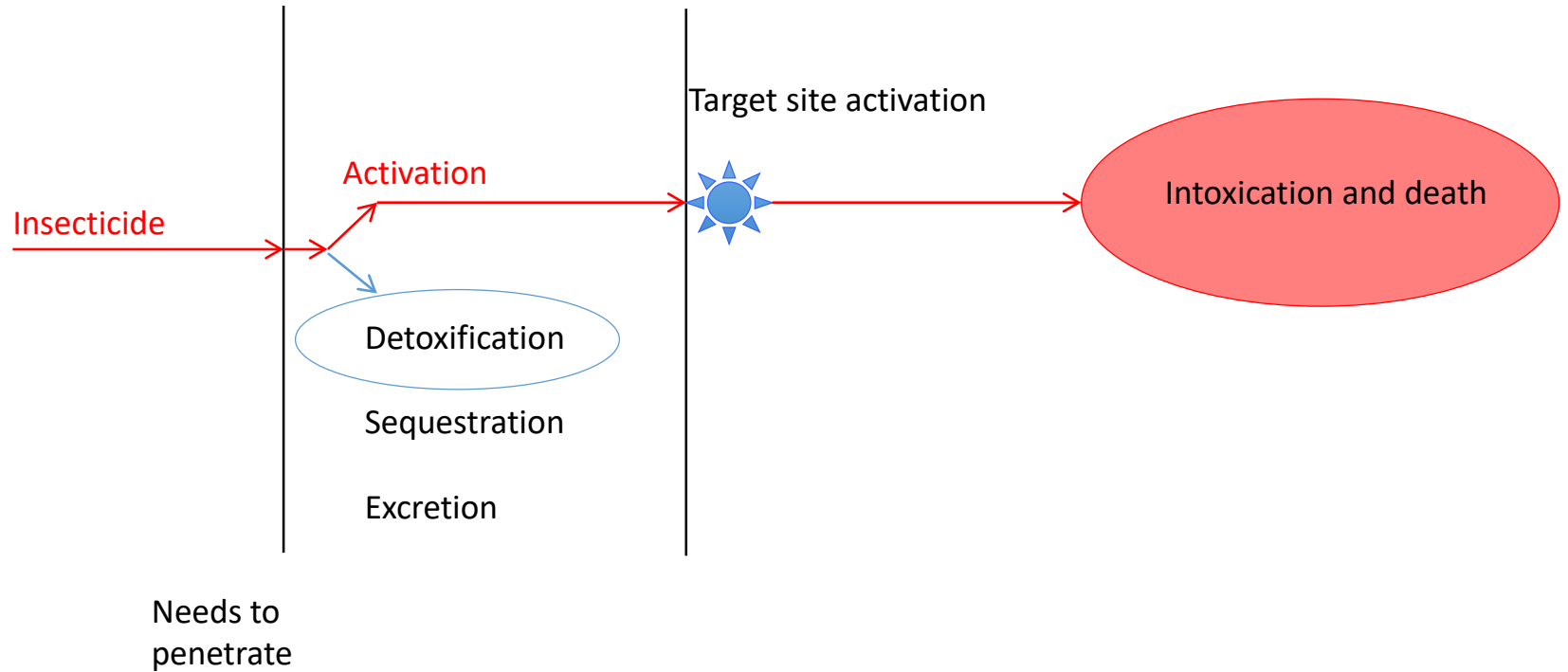
Persistence of resistance:

- When resistance takes a while to develop, it suggests R alleles are initially rare
- The rarity of R alleles suggests it comes with reduced fitness
- Thus, R alleles should fall to a very low frequency without insecticide selection
- Is this the case with ACP?: YES

Response of four field-sampled populations of ACP in Florida to imidacloprid and thiamethoxam after various intervals without exposure under laboratory rearing conditions.



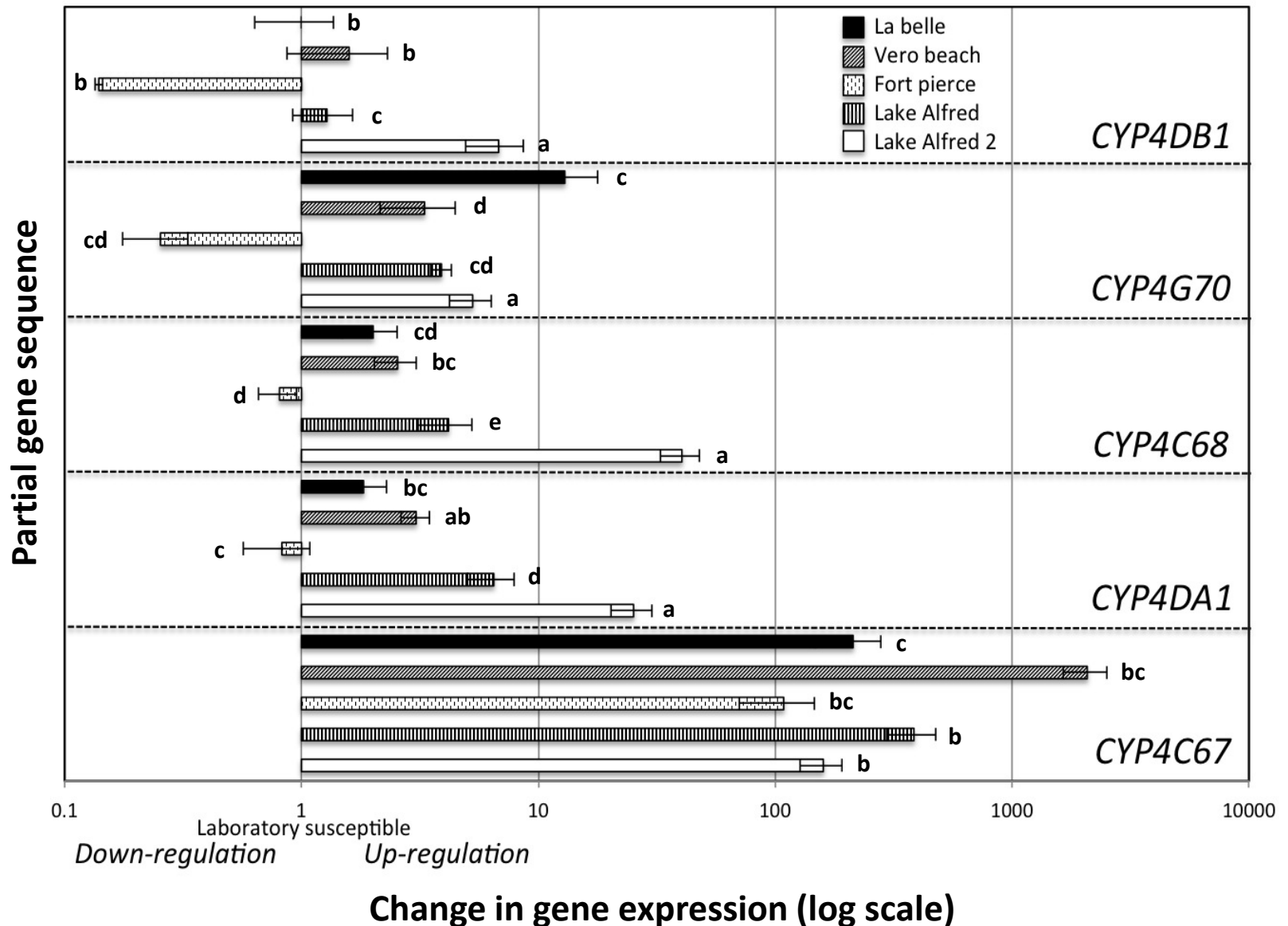
Resistance mechanisms: How does an insect become resistant?



Behavioral resistance-No current evidence



CYP4 gene expression upregulated in resistant populations throughout Florida



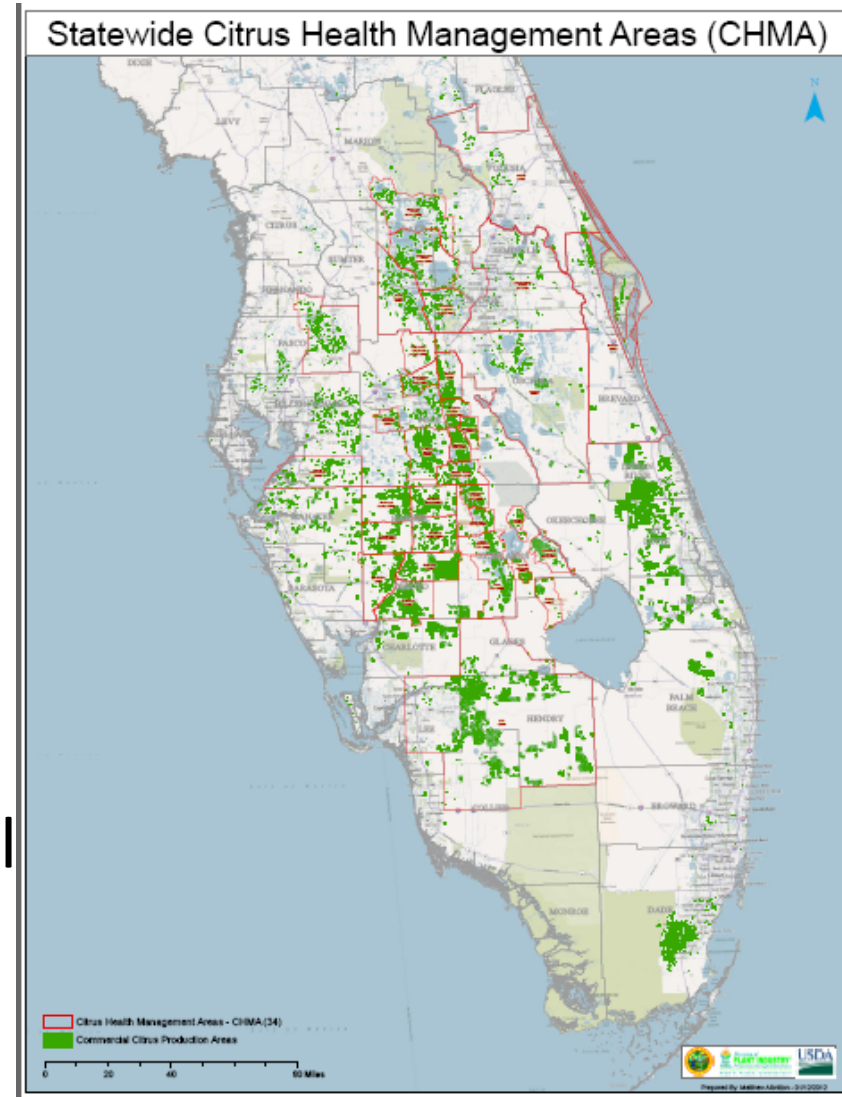
Increased Metabolism:

- The most common resistance mechanisms, called metabolic resistance. RR individuals have more enzymes or more efficient enzymes
- P450: Cytochrome P450 monooxygenases, involved in metabolism and resistance to all classes of insecticides
- Esterases: involved in resistance to organophosphates (OP), carbamates (Carb), and Pyrethroids
- GST: involved in resistance to DDT, OP and Py

Annual Survey to Monitor For Insecticide Resistance

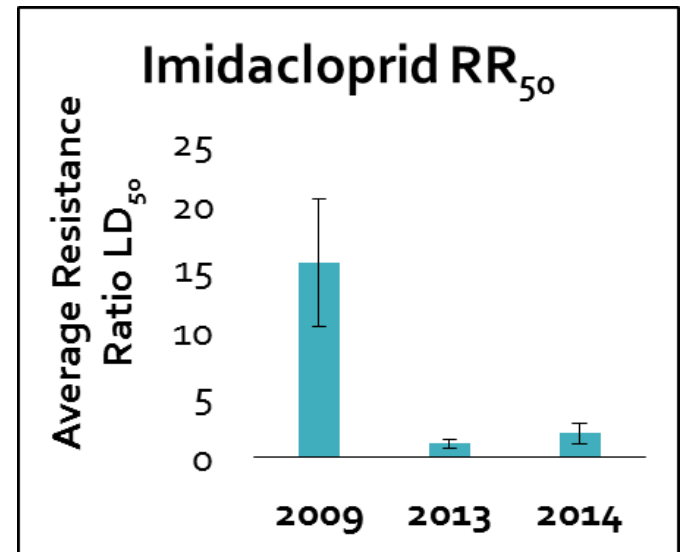
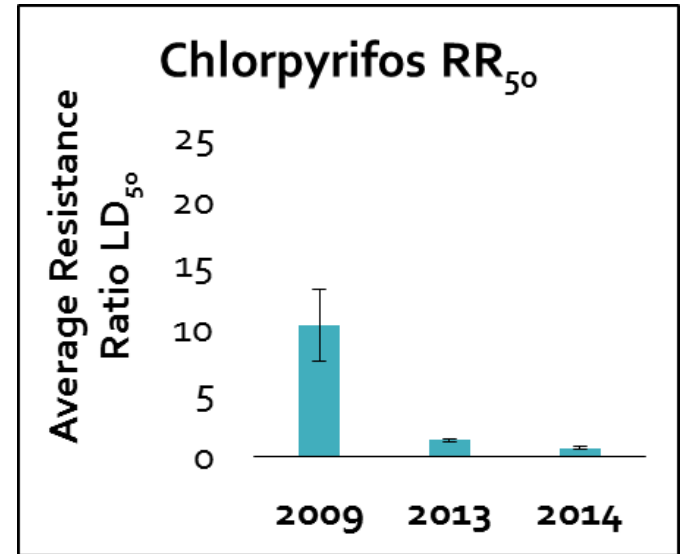
Four purposes:

1. Monitor for insecticide resistance in *D. citri* field populations
2. Study the changes that occur over time in the insecticidal response of *D. citri*
3. Determine the natural variation in *D. citri* insecticidal response
4. Refine spray schedules



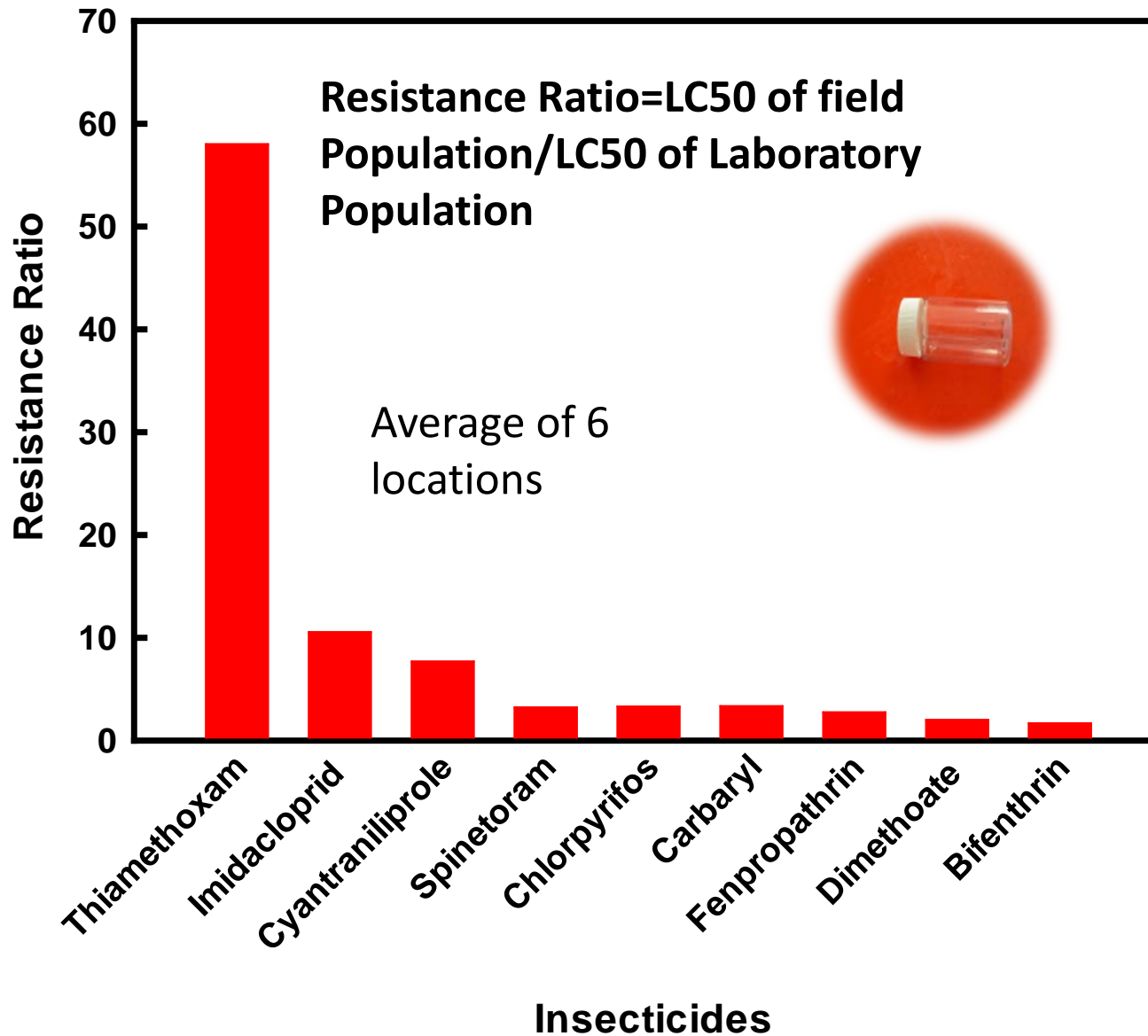
Average LD₅₀ Resistant Ratios

- RR_{50} = Field Population LD₅₀ / Laboratory Strain LD₅₀
- 2009: ratios were on the rise for some major classes of insecticides
- 2013 and 2014 fell back to susceptible levels
 - Hypothesis: Improved spray regimes through CHMA were working



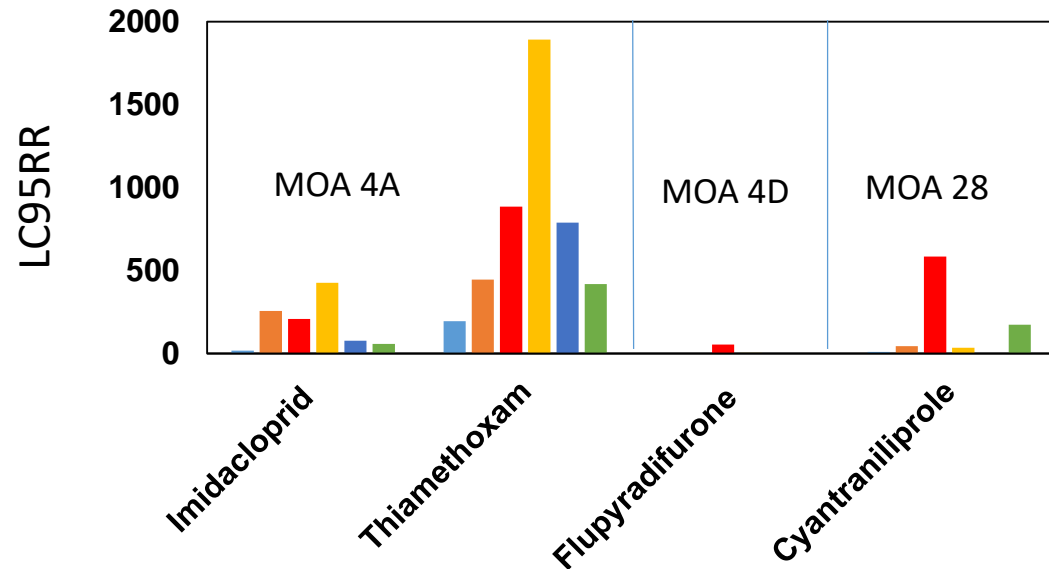
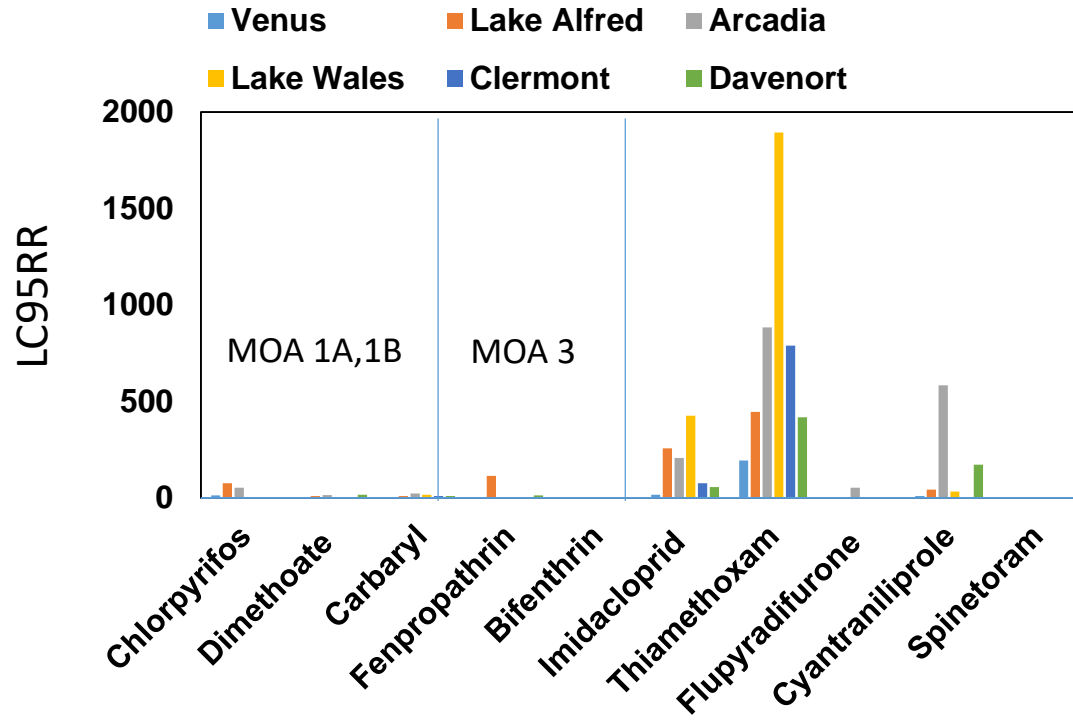
*Average of RR_{50} from all sites surveyed

Insecticide susceptibility—2017



2017 Resistance monitoring in 6 locations in Florida (LD95s)

| MOA | Class | Site of action |
|-----|-----------------|---|
| 1 | OPs & carbamate | AChE inhibitors Nerve action |
| 3 | pyrethroid | Sodium channel Nerve action |
| 4A | Neonicotinoid | Nicotinic AChR modulator Nerve action |
| 4D | butenolide | Nicotinic AChR modulator Nerve action |
| 28 | diamide | Ryanodine receptor modulator nerves/muscles |



'17-'18 Resistance monitoring in Florida (LD95s)

| Location/Insecticide | n | χ^2 | LD95 |
|----------------------|-----|----------|---------------------------|
| <u>Imidacloprid</u> | | | |
| Laboratory | 404 | 36.53 | 27.59 (11.34-91.00) |
| Clermont | 282 | 9.74 | 676.05 (36-363445) |
| Lake Alfred | 405 | 4.11 | 1108 (298-7005) |
| <u>Bifenthrin</u> | | | |
| Laboratory | 233 | 3.59 | 6.55 (2.41-29.43) |
| Clermont | 313 | 4.43 | 3.55 (1.33-14.04) |
| Lake Alfred | 267 | 5.94 | 3.17 (1.40-10.52) |
| <u>- Dimethoate</u> | | | |
| Laboratory | 263 | 9.23 | 21.08 (3.20-1788) |
| Clermont | 280 | 9.78 | 56.75 (8.51-4585) |
| Lake Alfred | 264 | 4.70 | 741 (150-8720) |

What do the textbooks say about all of this?

Insecticide resistance management (IRM) What can you do about resistance?

- Historically, growers have responded to resistance by
 - Increasing dosage and frequency of application which only accelerate the development of resistance
 - Switch to another insecticide---- so begins the pesticide treadmill
- In theory, resistance can be minimized by
 - Increasing survivorship of SS individuals
 - Decrease survivorship of resistant individuals (R allele)
- Three IRM strategies
 - Moderation
 - Saturation
 - Multiple attack

What do the textbooks say about all of this?

IRM by moderation:

- Strategy: Conserve susceptible alleles by reducing selection pressure in the following ways
 - higher threshold, low dosage, less frequent application
 - local, rather than area wide applications
 - Preserve refuge by treating only "hot spots" in a field
 - Use short residual insecticide and avoid slow-release formulation
 - Good for environment and natural enemies, but not practical for high value crops (fruits, veg.) and medical insects
- Not useful for Florida citrus.

What do the textbooks say about all of this?

Management by Saturation:

- Strategy: suppress resistant alleles by maximizing the mortality of R individuals in the following ways
 - High dosage
 - Apply at the most vulnerable stage of pests: age specific expression of resistance, low Resistance level at younger stages
 - Synergists to inhibit detoxification enzymes
 - Work if R allele is recessive and rare
 - Environmental concern
 - Necessary for high value crops/medical pests
- Some ideas might be useful, but likely not best approach to follow strictly

What do the textbooks say about all of this?

Management by multiple attack

- Strategy: suppress resistant alleles by diversifying selection forces and complicating pests' adaptation in the following ways
 - Mixed use of insecticides at low dosage that differ in mode of actions. A-resistant individuals can be killed by insecticide B and vice versa.
 - Insecticide rotation
 - Concern: cross resistance or multiple resistance
 - Necessary for high value crop/medical pests
- Some useful ideas here that we know can be implemented. Also, some useful ideas for further investigation.

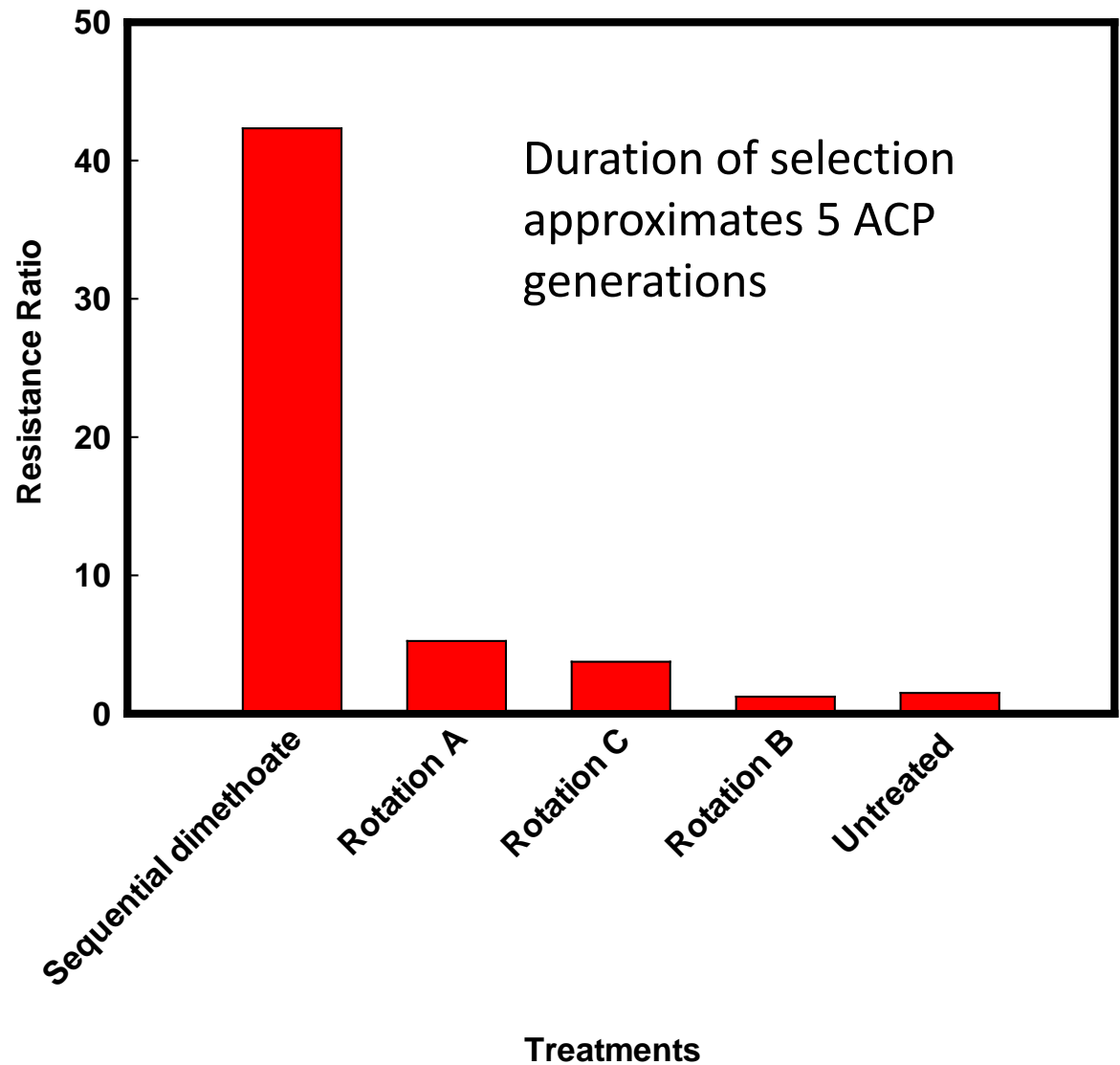
The best solution: Integrated Pest management (IPM)

- Combination of cultural, chemical and biological control tactics
- Diversifying selection forces and complicating adaptation
- Resistance to one tactics can be compensated for by other tactics

--But, as mentioned earlier, we don't yet have a full proof IPM system that prevents spread of HLB. So, we need to take what we've learned from the field and combine it with the current body of knowledge to manage resistance. And, there is plenty of evidence that we are able to do it.

Rotation is effective-
experimentally
verified

Susceptibility of ACP
following 5
consecutive
dimethoate sprays
versus various
insecticide rotations

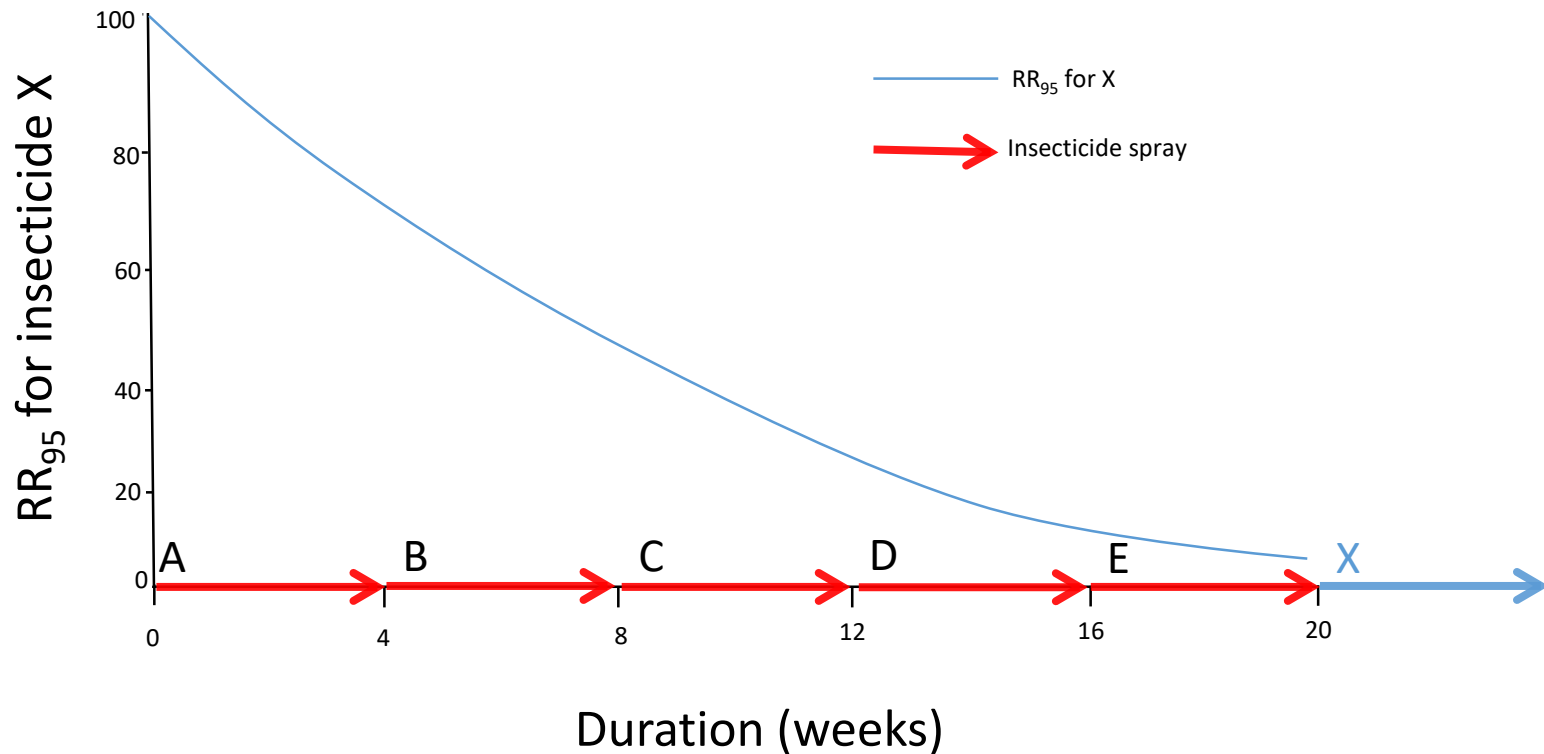


Rotation A: organophosphate, microbial, synthetic pyrethroid, insect growth regulator and neonicotinoid;
Rotation B: neonicotinoid, synthetic pyrethroid, microbial, organophosphates and insect growth regulator;
Rotation C: microbial, insect growth regulator, organophosphate, neonicotinoid and synthetic pyrethroid;
Sequential dimethoate: 5 sprays with dimethoate (Resistance Ratio=LC50 of field Pop/LC50 of Laboratory Pop).

Rotating 5 modes of action in sequence could also theoretically cause reversal to susceptibility for over-selected MOAs—needs verification

5 sequential MOAs-Hypothetical situation if known resistance to insecticide X

5 sprays every 4 weeks under ideal conditions is close to 5 ACP generations



If you were doing young tree care without soil application of neonicotinoids:

- Foliar sprays may serve as replacements for soil drenches as warranted
- Insecticide alternatives such as Surround reduce ACP numbers and help prevent pathogen inoculation
- Reflective mulches further reduce ACP populations

| Tree size | Jan | Feb | Mar | Apr | May | Jun | Jul | Aug | Sep | Oct | Nov | Dec |
|--|------------------------------|------|------|------|------|------|------|------|------|------|------|------|
| Non-bearing trees Resent -5 year < 3'-9' | FS-1 | FS-2 | FS-3 | FS-4 | FS-5 | FS-1 | FS-2 | FS-3 | FS-4 | F-5S | FS-1 | FS-2 |
| | | | | | | | | | | | | |
| | Season-long Reflective mulch | | | | | | | | | | | |
| FS= Foliar Insecticide Spray; Green shaded areas indicate opportunity for Kaolin clay application. Rate 25 lbs/100 gal of water works well. Can cover more than an acre. Can interfere with translaminar insecticides* | | | | | | | | | | | | |

Do we have at least 5 modes of action to rotate?

| MOA | Class | Site of action | |
|-----|--------------------|---|---|
| 1 B | Ops | AChE inhibitors Nerve action | Lorsban 4 E, Dimethoate 4 E, Imidan 70 W |
| 3A | pyrethroid | Sodium channel Nerve action | Baythroid XL, Danitol 2.4 EC, Mustang |
| 4A | Neonicoti- noid | Nicotinic AChR modulator Nerve action | Various imidacloprid products, 2F, 4F, 4.6F; Actara (foliar), Platinum 75 G (drench) Belay (drench) |
| 4D | butenolide | Nicotinic AChR modulator Nerve action | Sivanto 200 SL |
| 28 | diamide | Ryanodine receptor modulator nerves/muscles | Exirel (foliar), Verimark (drench) |

Others? Delegate WG (5), Movento (23), Apta and Portal (21A), Oil (sufficant/deterrent)

Does order of the sequence matter?

| Treatment | May | June | July | September | October |
|-------------------|-----------------------------|-----------------------------|------------------------------|---------------|---------------|
| Rotation A | dimethoate | abamectin + thiamethoxam | fenpropathrin + abamectin | diflubenzuron | imidacloprid |
| Rotation B | imidacloprid abamectin + | fenpropathrin | abamectin + thiamethoxam | dimethoate | diflubenzuron |
| Rotation C | thiamethoxam | diflubenzuron | dimethoate | imidacloprid | fenpropathrin |

No

Diversify Chemical Use

Multiple targets

Adults/nymphs

Secondary pests

Rotation

Reduce Frequency

and use selective MOAs

when possible

Cost

Dormant or Border

Growing season/whole block

| Months | Nov-Dec | Jan | Feb-Mar | Apr | May - June | July - Aug | Sep-Oct |
|--|---|---|--|---|--|---|---|
| Products | OP ¹³ (e.g. Imidan, Dimethoate, chlorpyrifos) | Pyrethroid ¹⁴ (Mustang Danitol Baythroid) | *Sivanto ⁹ *Movento ¹ *Portal ² *Micromite ⁴ *Intrepid ⁵ Exirel ⁶ | OIL ⁷ Portal ² Micromite ⁴ Exirel ⁶ Apta ⁸ Sivanto ⁹ | Movento ¹ Delegate ¹⁰ Abamectin ¹¹ Knack ¹² Exirel ⁶ Apta ⁸ Sivanto ⁹ | Oil ⁷ Sivanto ⁹ Apta ⁸ OP ¹³ | Movento ¹ Delegate ¹⁰ Apta ⁸ Sivanto ⁹ |
| Pests | ACP Weevils | ACP Weevils | ACP Mites Leafminer Weevils Scales Aphids | ACP Mites Leafminer Weevils Aphids | ACP Rustmite Leafminer Scales | ACP | ACP Rustmite Leafminer |
| ACP+++^{1,6,8,9,13,14} ACP++^{2,10} ACP+^{4,7,11} Leafminer^{4,5,6,10} Rustmite^{1,4,7,8,11} Scales^{1,12} Aphids⁹ Mealybugs¹ | | | | | | | |

Cultural control can help: Repellents, Attractants, Barriers – **comments/questions?**

- **Reflective mulches and kaolin clay** help repel psyllids, in combination with insecticides, especially in newly planted orchards.
- **Windbreaks** are helpful in reducing psyllids and planted trees or artificial screens could be part of a ‘systems’ approach
- **Reset plantings** have fewer psyllids than replants of entire orchards
- Trees enclosed in **protective structures** would be the ultimate barrier to psyllids

**Biological control (if present) could help too.
We need to figure out its effect on IRM and if
we can boost it**

Stansly, Setamou, Martini, others



Conclusions:

- ACP in Florida show insecticide resistance in numerous locations and regions
- Soil application of neonicotinoids causes persistent sub-lethal dosages of active ingredient in plants that exacerbates resistance
- Insecticide resistance can be effectively managed by rotating 5 modes of action in sequence
- The sequence in which you apply MOAs in a rotation does not affect IRM of ACP
- Six ACP generations are required to for reversal of insecticide-resistant populations to susceptibility