

Quarter 2 (7/1/2019 – 9/30/2019) Report

Florida Department of Health Contract CODQJ

Improving our understanding of domestic mosquito control of *Aedes aegypti*, *Ae. albopictus*,
and *Culex quinquefasciatus* through assessments of insecticide susceptibility

Prepared by:

PI: Eva Buckner, PhD¹

Co-PI: Barry Alto, PhD²

Daviela Ramirez

University of Florida, IFAS

Florida Medical Entomology Laboratory

200 9th St SE

Vero Beach, FL 32962

¹eva.buckner@ufl.edu 772-226-6606

²bwalto@ufl.edu 772-226-6630

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Table of Contents

Task List.....3

Tasks Progress.....5

Task List

1. Deploy traps each quarter to collect a minimum of 100 eggs from adult *Aedes* and *Culex* mosquitoes or collect 100 *Aedes* and *Culex* larvae from within the identified sites or collect eggs from adult *Aedes* and *Culex* mosquitoes hatched from previously collected eggs from identified sites that were reared to adulthood and allowed to blood feed and lay eggs. Document the number of eggs and larvae collected and the number and the species of adult mosquitoes that hatched from collected eggs in the Quarterly Report.
2. Conduct insecticide resistance testing on mosquitoes collected as eggs (parental generation) or successive generations (within two generations of parental generation) of mosquitoes within 60 days of collection and hatching of eggs. Document the insecticide resistance testing and results in the Quarterly Report.
3. Conduct CDC bottle bioassay testing on mosquitoes from a minimum of three identified sites against one pyrethroid and one organophosphate each quarter. Document the bioassay testing and results in the Quarterly Report and post the CDC bottle bioassay results to Provider's reporting website, <https://fme1.ifas.ufl.edu/>.
4. Map the distribution of where *Aedes* and *Culex* eggs or larvae are collected and used in CDC bottle bioassays each quarter. Document the mapped distribution in the Quarterly Report.
5. Distribute the results of the CDC bottle bioassay testing to the Florida Mosquito Control Program managers in the counties of the identified sites each quarter. Document the distribution of the results in the Quarterly Report.
6. Prepare a Quarterly Report, post it on Provider's reporting website, and submit it to the Contract Manager within 15 days following the end of each quarter, but no later than invoice submission. At a minimum, include the following information in the report:
 - a. The number of eggs and larvae from adult mosquitoes collected and hatched;
 - b. The number of eggs and larvae from adult mosquitoes in which insecticide testing was conducted;
 - c. Documentation of mapping of egg collection results including species identification and location of collection;
 - d. Documentation of results of CDC bottle bioassay testing of mosquitoes for insecticide resistance; and
 - e. Documentation of CDC bottle bioassay testing results distribution to Florida Mosquito Control Program Managers.
7. Identify the methods for distributing information on resistance to tested insecticide active ingredients. Prepare an Annual Report, including the identified methods, and submit it to the Contract Manager within 45 days from the end of the contract term, but no later than submission of the final invoice. At a minimum, include the following in the report:

- a. The method for informing Florida Mosquito Control Programs, the Department, and the general public on the regions of Florida that are most likely to have populations of Zika, Dengue, Chikungunya, and West Nile Virus mosquito vectors;
- b. The method for informing Florida Mosquito Control Programs and the Department on the efficacy of the insecticides that are currently being used in their programs (i.e., whether the chemicals are working as they should to reduce the mosquito populations); and
- c. The method for informing Florida Mosquito Control Program managers on the relationship between the CDC bottle bioassay (a lab assay) and the efficacy of spraying mosquitoes at the insecticide label rates.

Tasks Progress

1. Traps were deployed this quarter, and 732 *Aedes* eggs and 147 *Culex quinquefasciatus* rafts were collected from the traps. 2,420 *Culex quinquefasciatus* larvae were also collected. Of the 732 F1 *Aedes* eggs collected, there was an 86% hatch rate, resulting in 703 F2 *Aedes* adults emerging. The *Culex quinquefasciatus* larvae collected had an 83% survivorship rate, resulting in 12,200 *Culex quinquefasciatus* adults emerging. The species identification for the resulting *Aedes* adult mosquitoes was *Aedes aegypti*. The *Aedes* eggs processed this quarter came from the following county and site within the county:

- a. Pinellas County
 - i. Sawgrass

The *Culex quinquefasciatus* rafts and larvae processed this quarter came from the following counties and sites within those counties:

- a. Bay County
 - ii. Gulf
 - iii. Hutchinson
 - iv. Nautilus
- b. Brevard County
 - i. Viera
- c. Hernando County
 - i. Spring Hill
- d. Lake County
 - i. Bloxham
 - ii. Neely
- e. Martin County
 - i. 138th Street
- f. Miami-Dade County
 - i. Mosquito Control
- g. Pinellas County
 - i. Lafayette
- h. Polk County
 - i. Ben Durrance
- i. Seminole County
 - i. Cameron
 - ii. Cascade

2. Since the end of the previous quarter, 3 populations of *Aedes aegypti*, 2 populations of *Ae. albopictus*, and 14 populations of *Culex quinquefasciatus*, respectively, have been tested for insecticide resistance using the CDC bottle bioassay. Based on the mortality

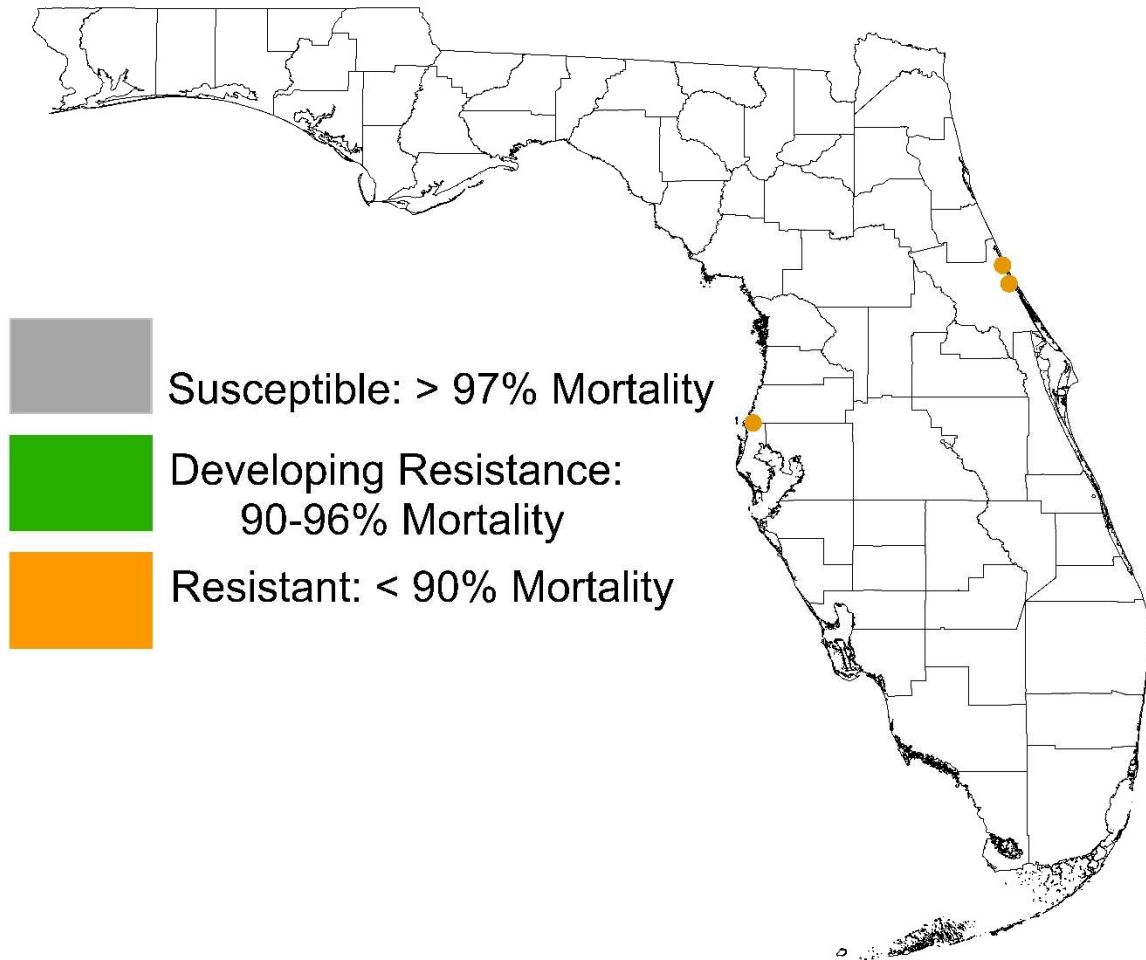
observed at a diagnostic time, the mosquito populations were classified as susceptible, developing resistance, or resistant to the various active ingredients. These results were incorporated into maps and can be found on pages 7 - 24 of this report.

3. CDC bottle bioassay testing was performed on mosquitoes from 19 sites against at least one pyrethroid (permethrin) and one organophosphate (malathion) active ingredient (AI). The total number of active ingredients that each mosquito population was tested against can be found in the table below. The CDC bottle bioassay results for each active ingredient can also be found at <https://fmeal.ifas.ufl.edu/>.

Species Tested	County	Site	# of AIs Tested
<i>Aedes aegypti</i>	Volusia	Nova	6
<i>Aedes aegypti</i>	Volusia	YMCA	6
<i>Aedes aegypti</i>	Pinellas	Sawgrass	6
<i>Aedes albopictus</i>	Escambia	Amanda Lane	6
<i>Aedes albopictus</i>	Volusia	Nova	6
<i>Culex quinquefasciatus</i>	Bay	Gulf	6
<i>Culex quinquefasciatus</i>	Bay	Hutchinson	6
<i>Culex quinquefasciatus</i>	Bay	Nautilus	6
<i>Culex quinquefasciatus</i>	Brevard	Viera	6
<i>Culex quinquefasciatus</i>	Hernando	Spring Hill	6
<i>Culex quinquefasciatus</i>	Lake	Bloxham	6
<i>Culex quinquefasciatus</i>	Lake	Neely	6
<i>Culex quinquefasciatus</i>	Martin	138th Street	6
<i>Culex quinquefasciatus</i>	Miami-Dade	Mosquito Control	6
<i>Culex quinquefasciatus</i>	Monroe	Key West	5
<i>Culex quinquefasciatus</i>	Pinellas	Lafayette	6
<i>Culex quinquefasciatus</i>	Polk	Ben Durrance	6
<i>Culex quinquefasciatus</i>	Seminole	Cameron	6
<i>Culex quinquefasciatus</i>	Seminole	Cascade	6

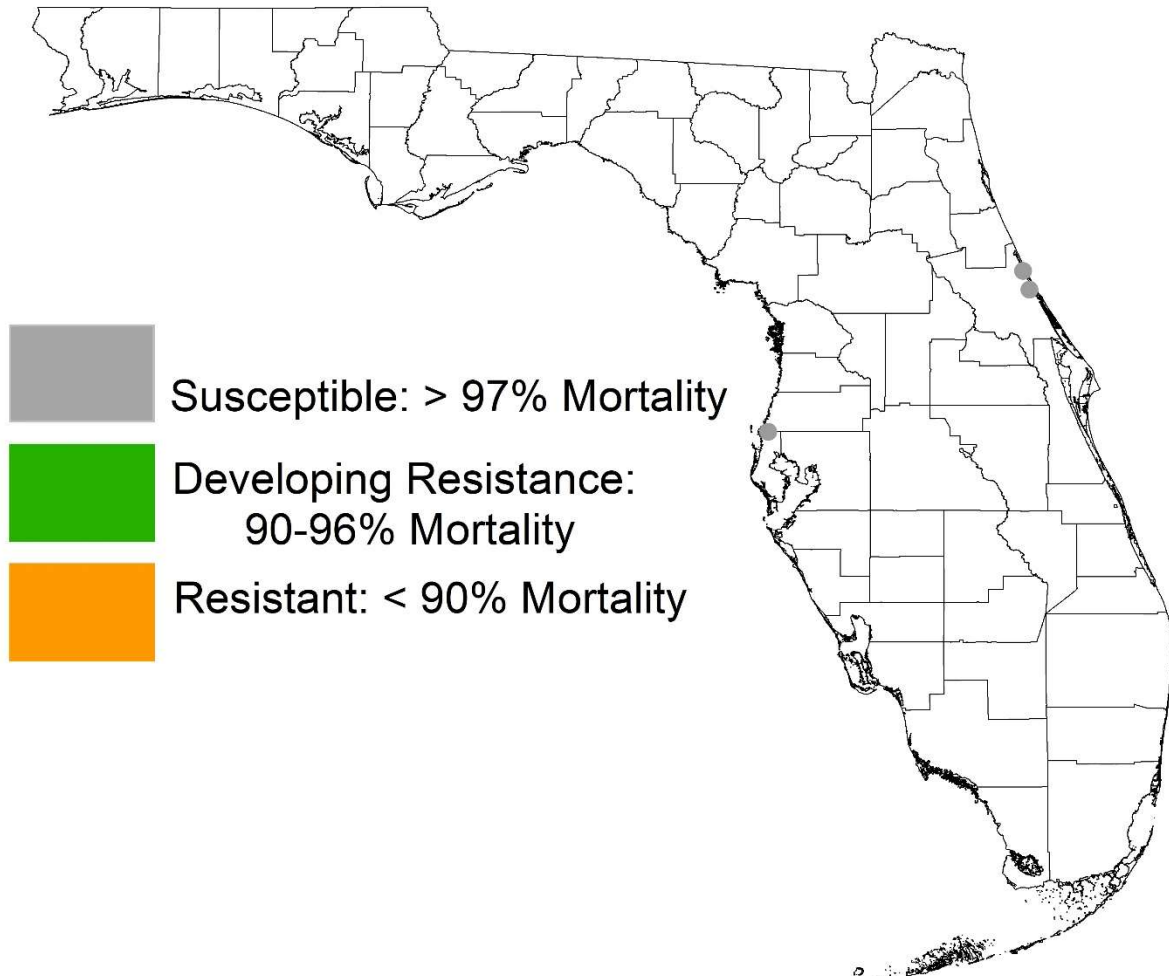
4. The distribution of where *Aedes* and *Culex* eggs or larvae were collected from and used in CDC bottle bioassays was mapped and can be found on page 25 of this report. The distribution map can also be found at <https://fmeal.ifas.ufl.edu/>.
5. Results of the CDC bottle bioassay testing were distributed as reports by email to the Florida Mosquito Control Program managers in the counties with sites tested this quarter. These emails were forwarded to the Contract Manager. An example report can be found on pages 26 - 27.
6. This Quarterly Report in combination with the information posted on the Reporting website at <https://fmeal.ifas.ufl.edu/> satisfies this task.
7. All necessary information will be provided in the Final Annual Report.

Species: *Aedes aegypti*
Active Ingredient: permethrin



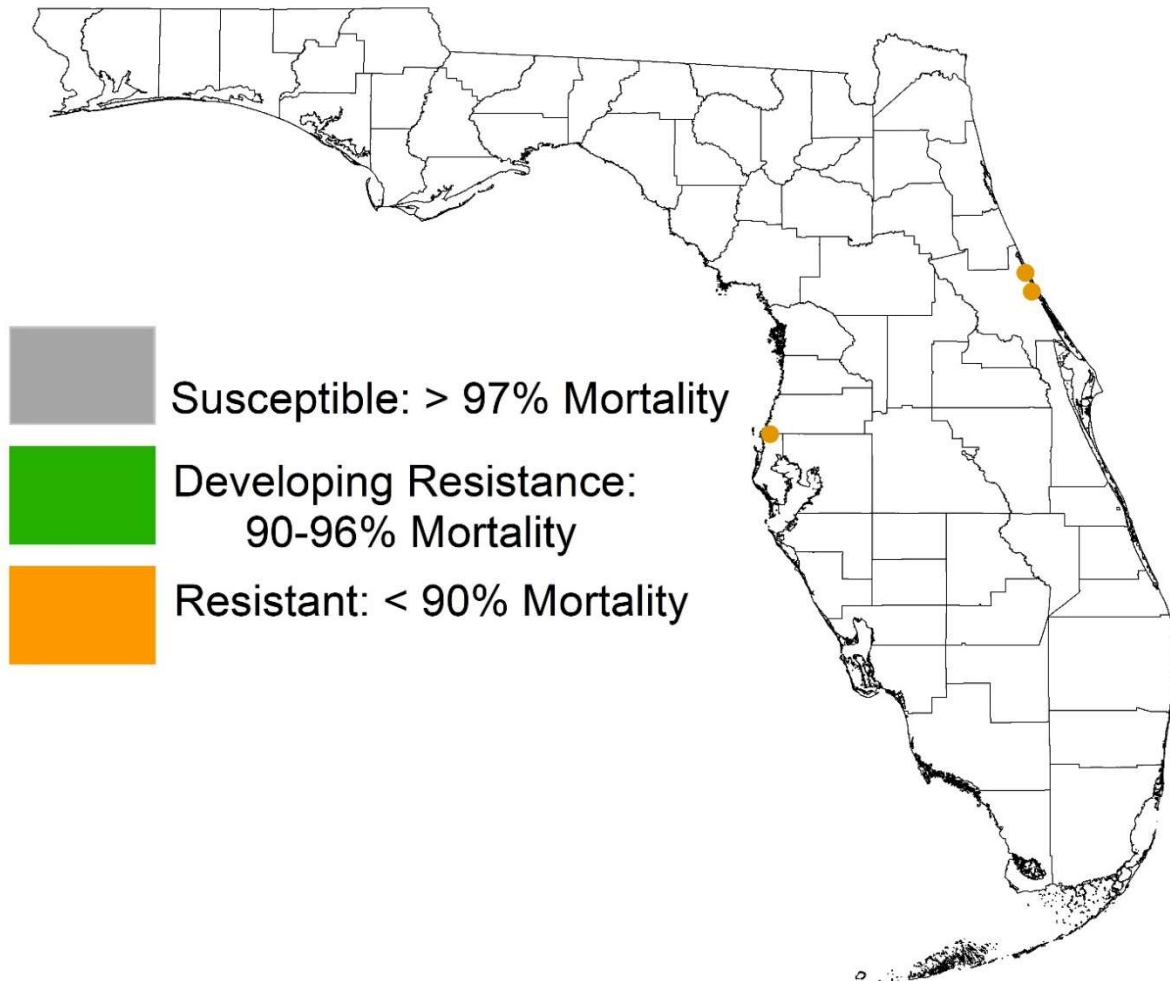
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Species: *Aedes aegypti*
Active Ingredient: malathion



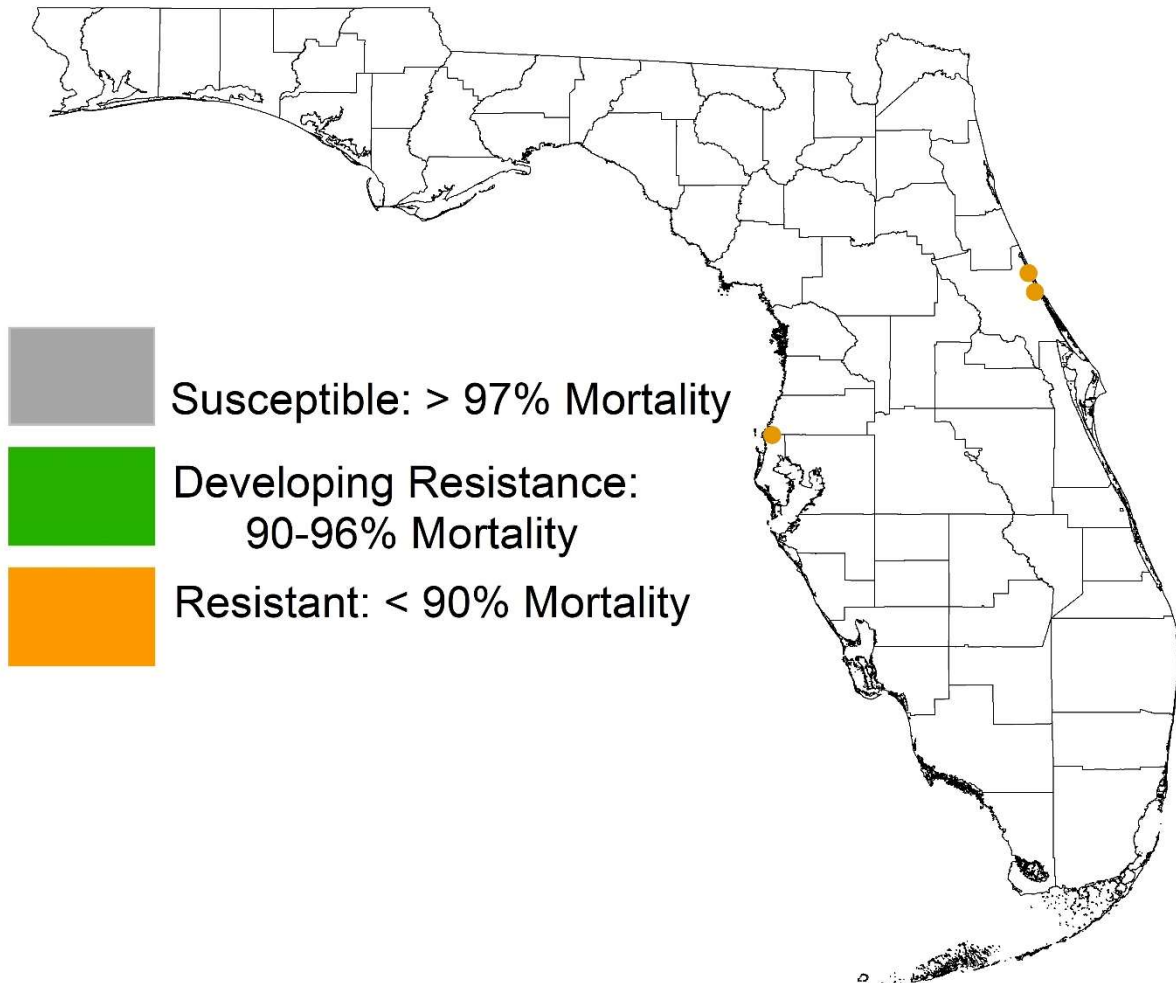
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
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Species: *Aedes aegypti*
Active Ingredient: naled



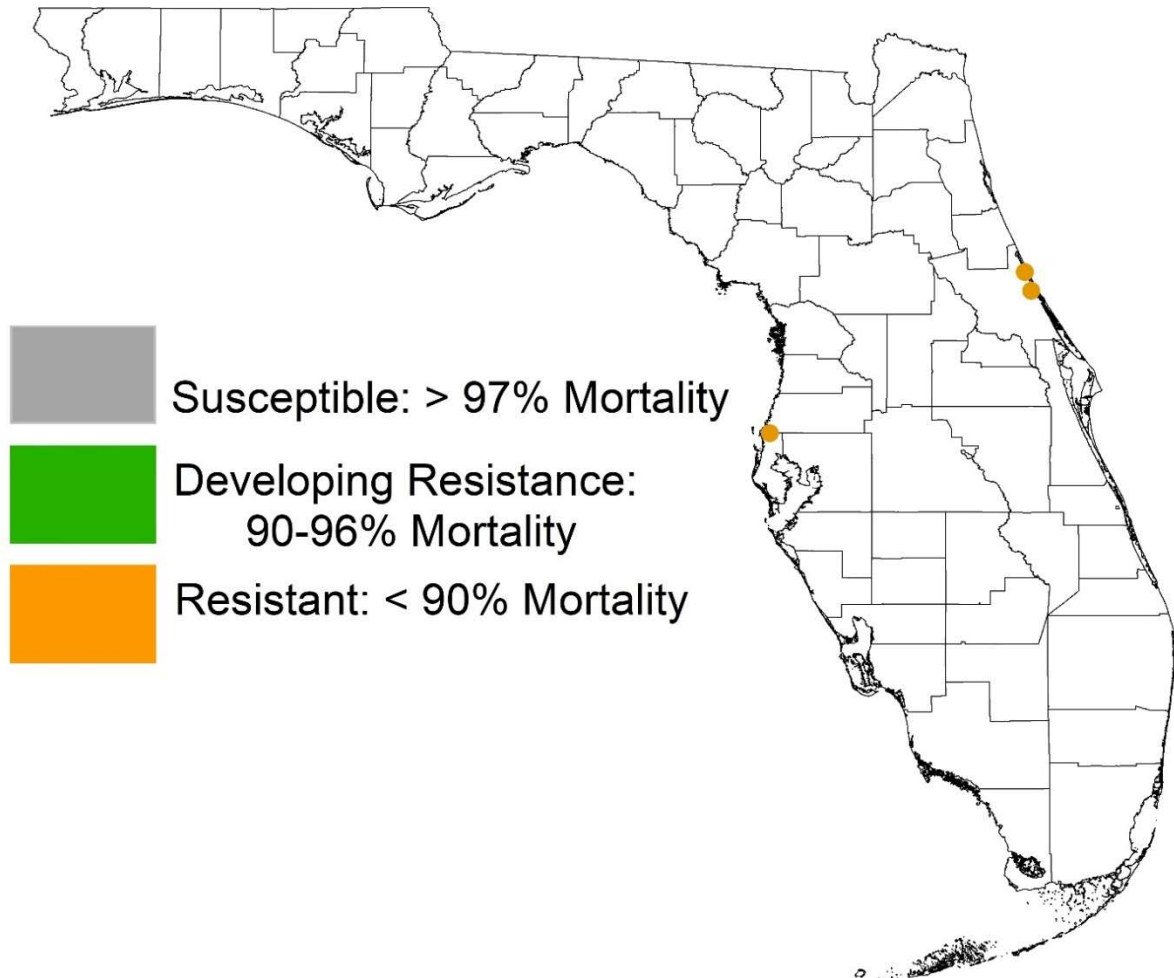
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
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Species: *Aedes aegypti*
Active Ingredient: deltamethrin



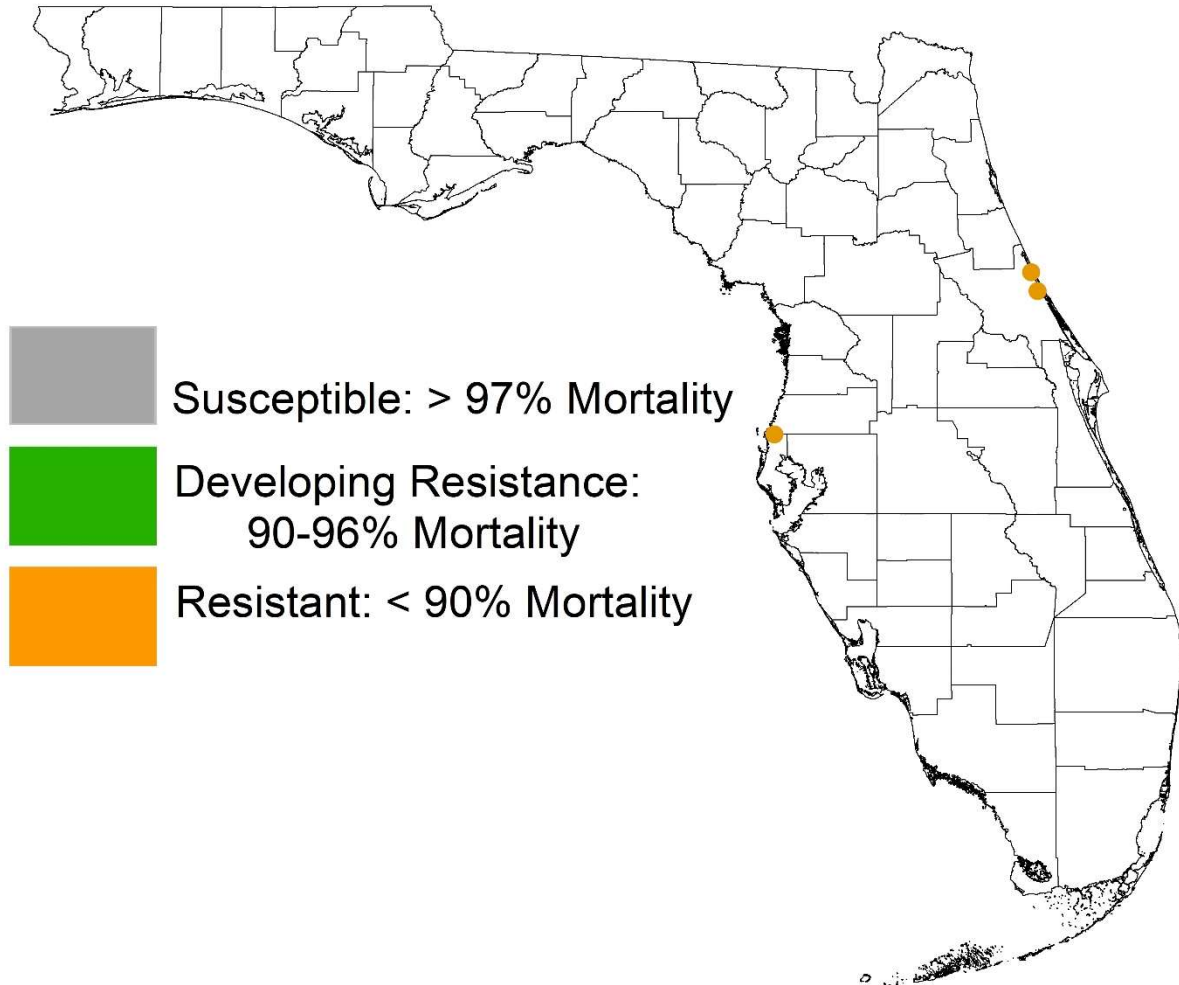
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
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Species: *Aedes aegypti*
Active Ingredient: etofenprox



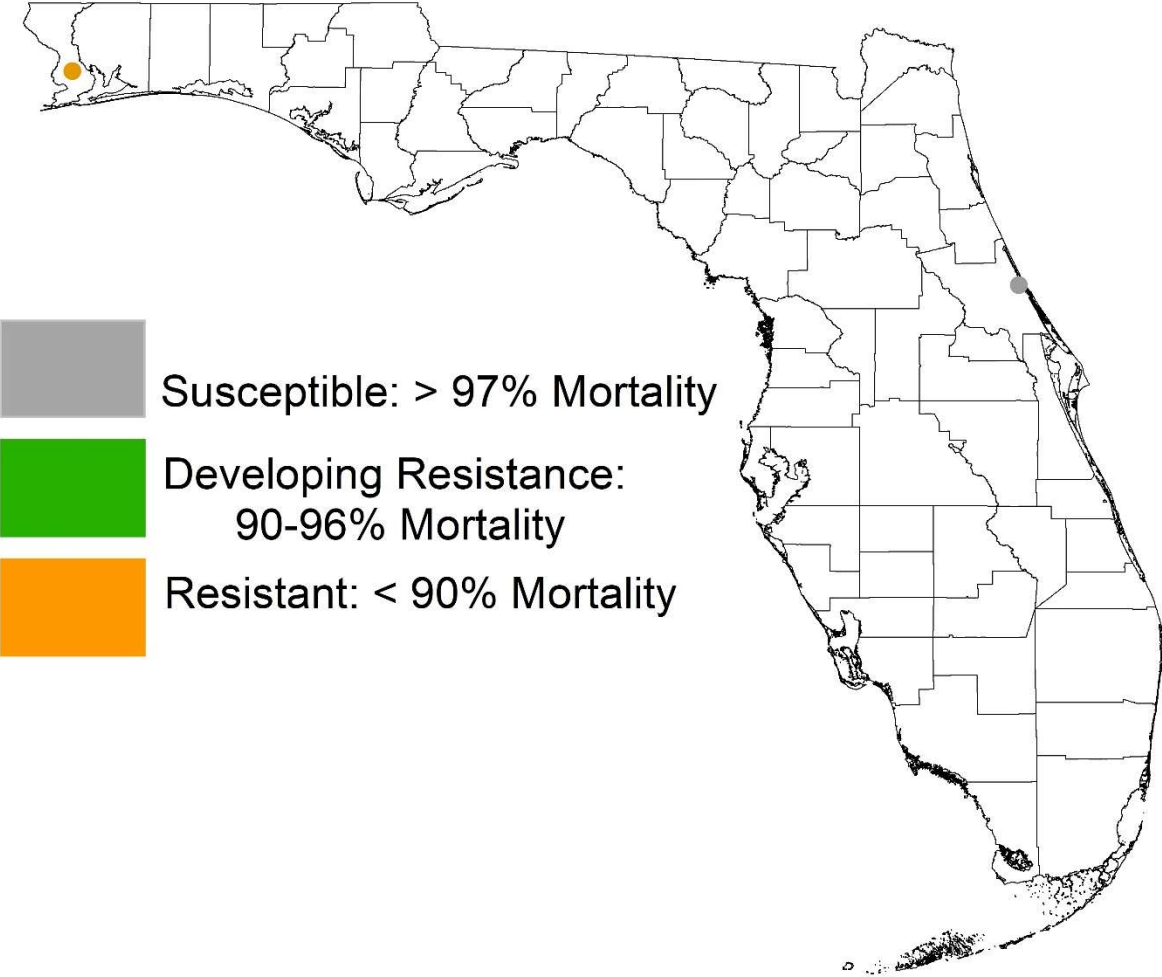
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Species: *Aedes aegypti*
Active Ingredient: sumithrin



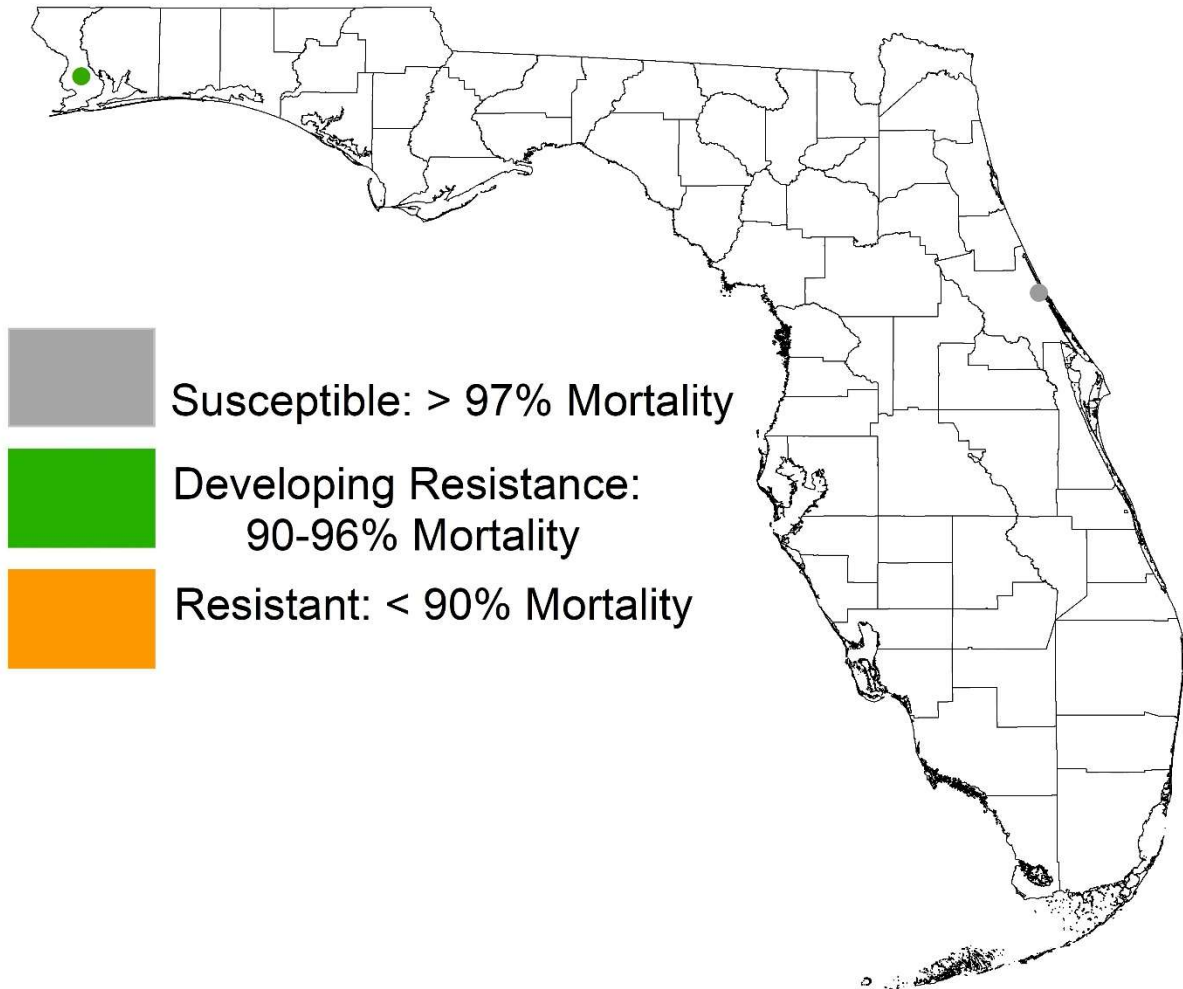
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
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Species: *Aedes albopictus*
Active Ingredient: permethrin



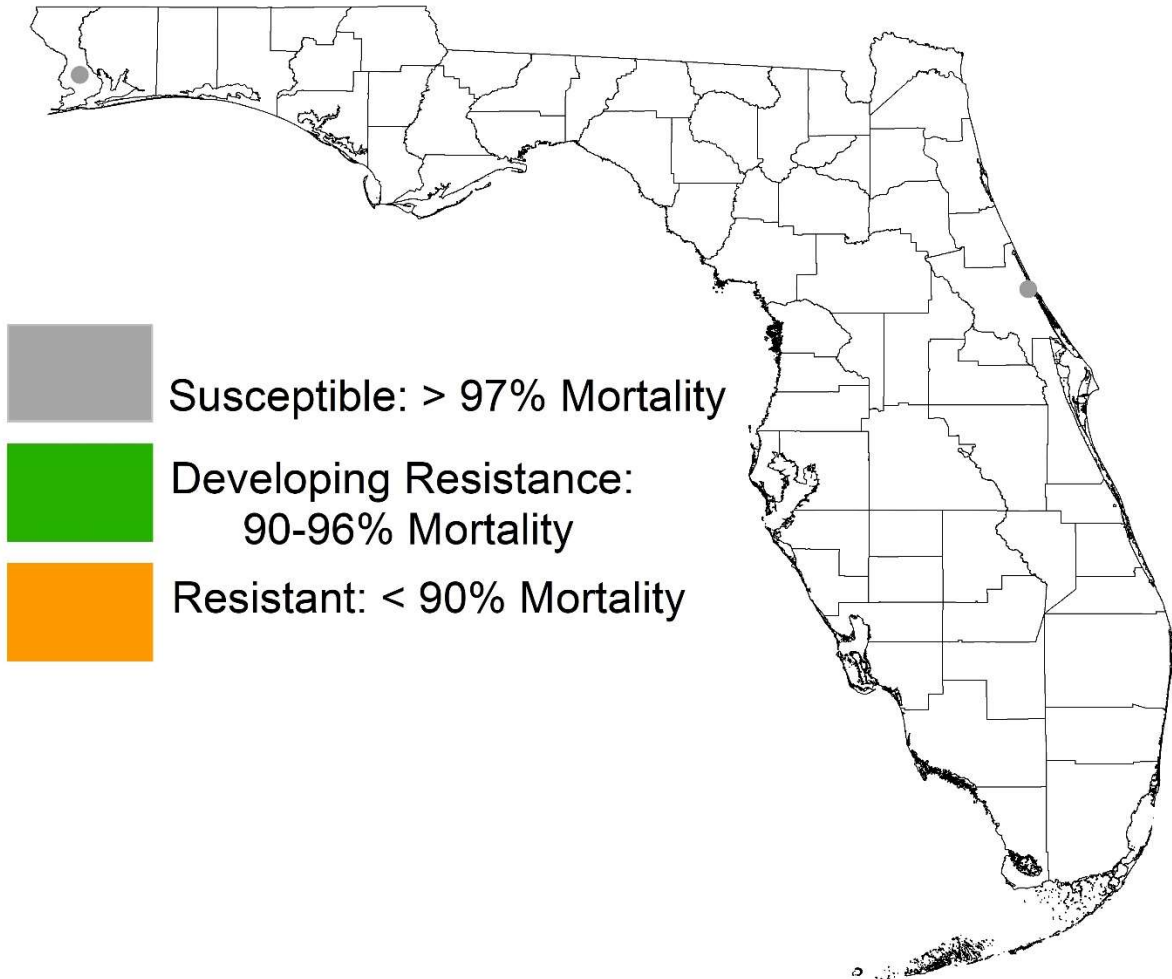
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Species: *Aedes albopictus*
Active Ingredient: malathion



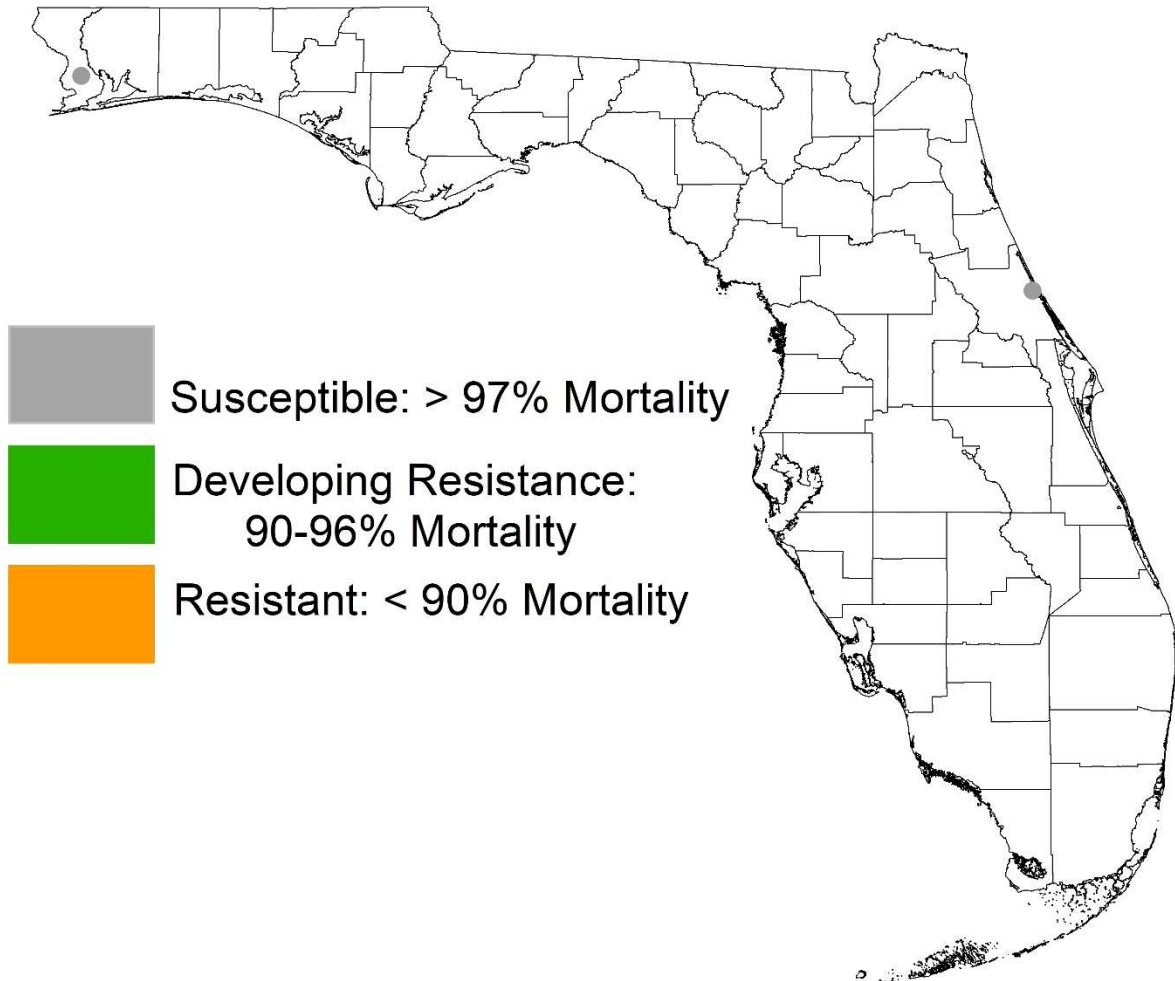
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Species: *Aedes albopictus*
Active Ingredient: naled



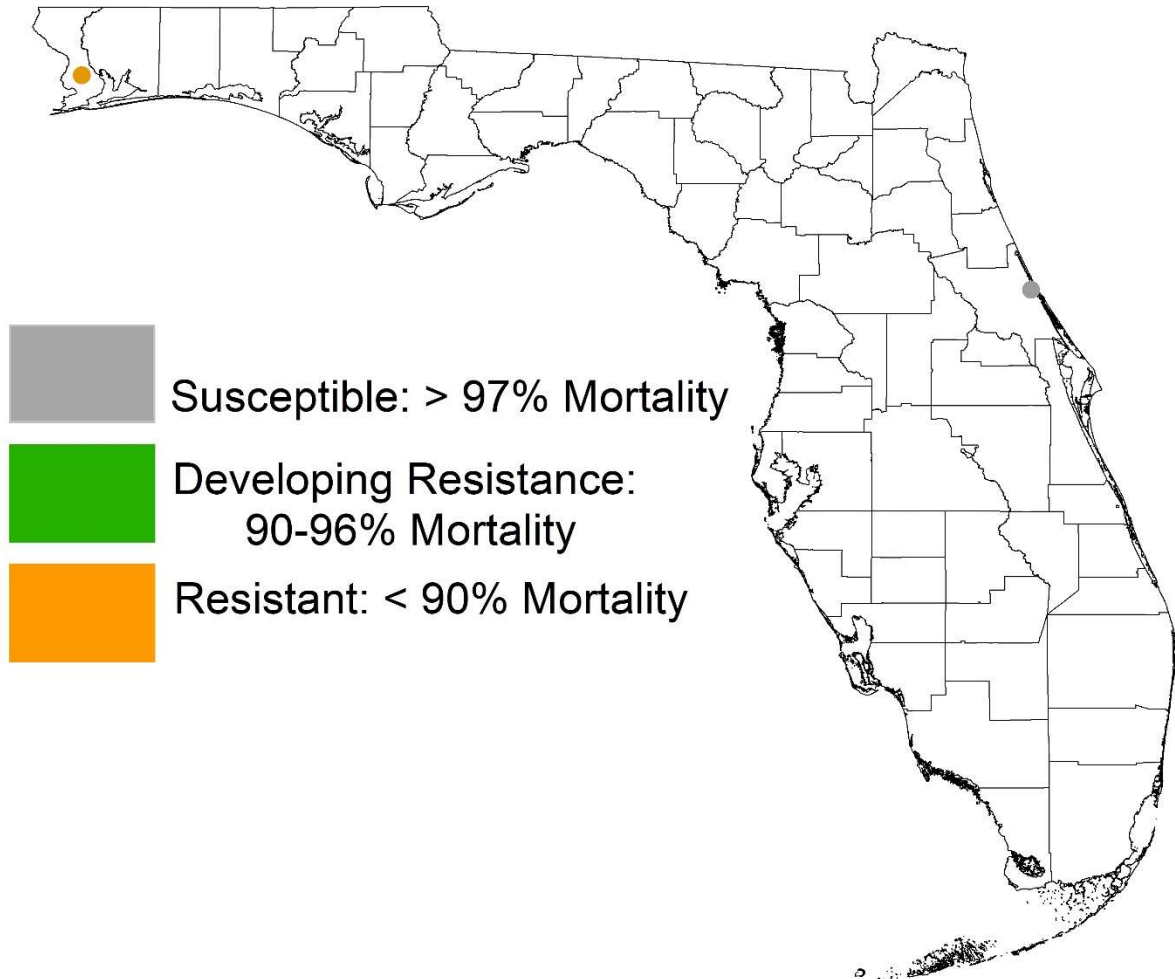
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Species: *Aedes albopictus*
Active Ingredient: deltamethrin



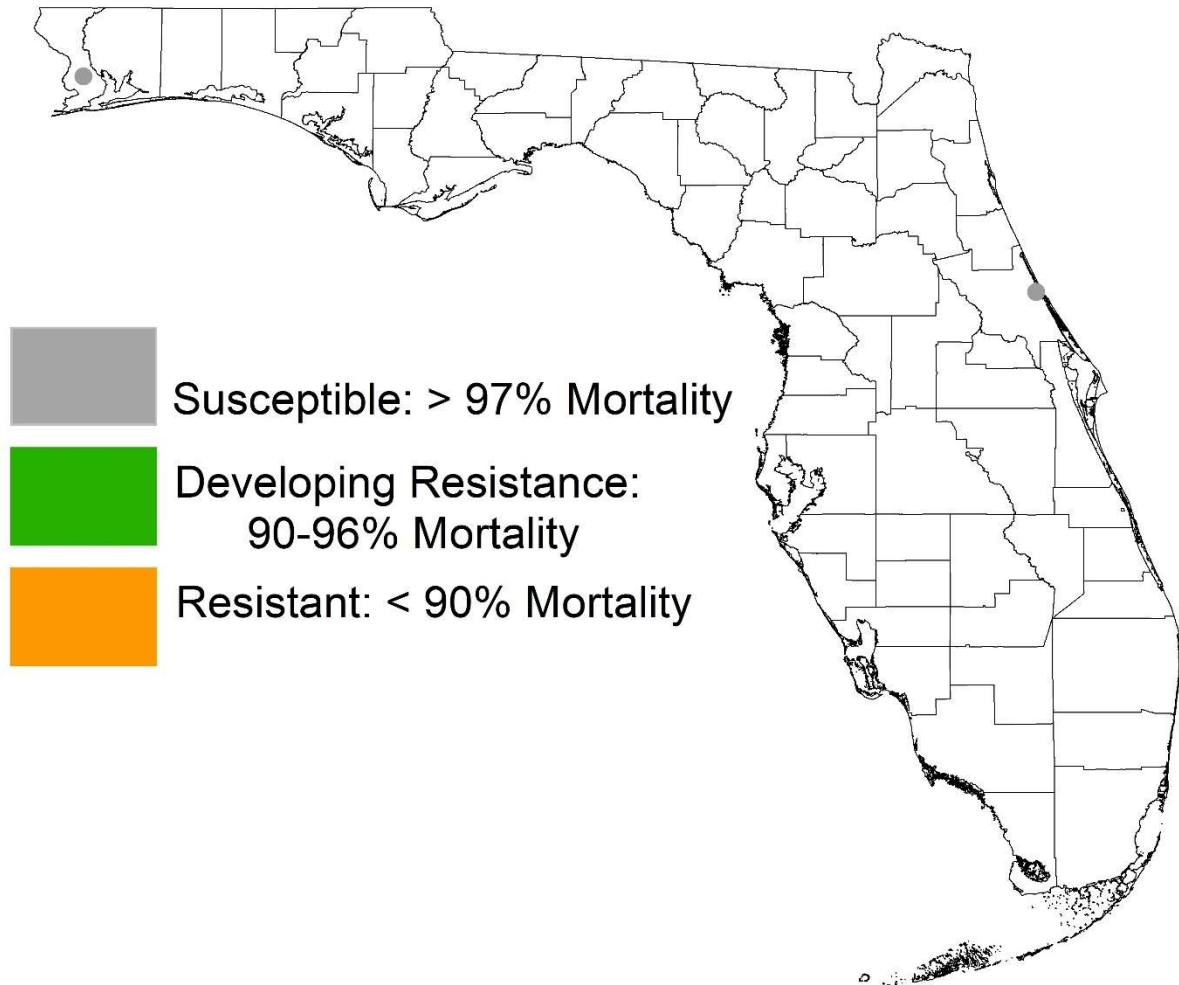
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Species: *Aedes albopictus*
Active Ingredient: etofenprox



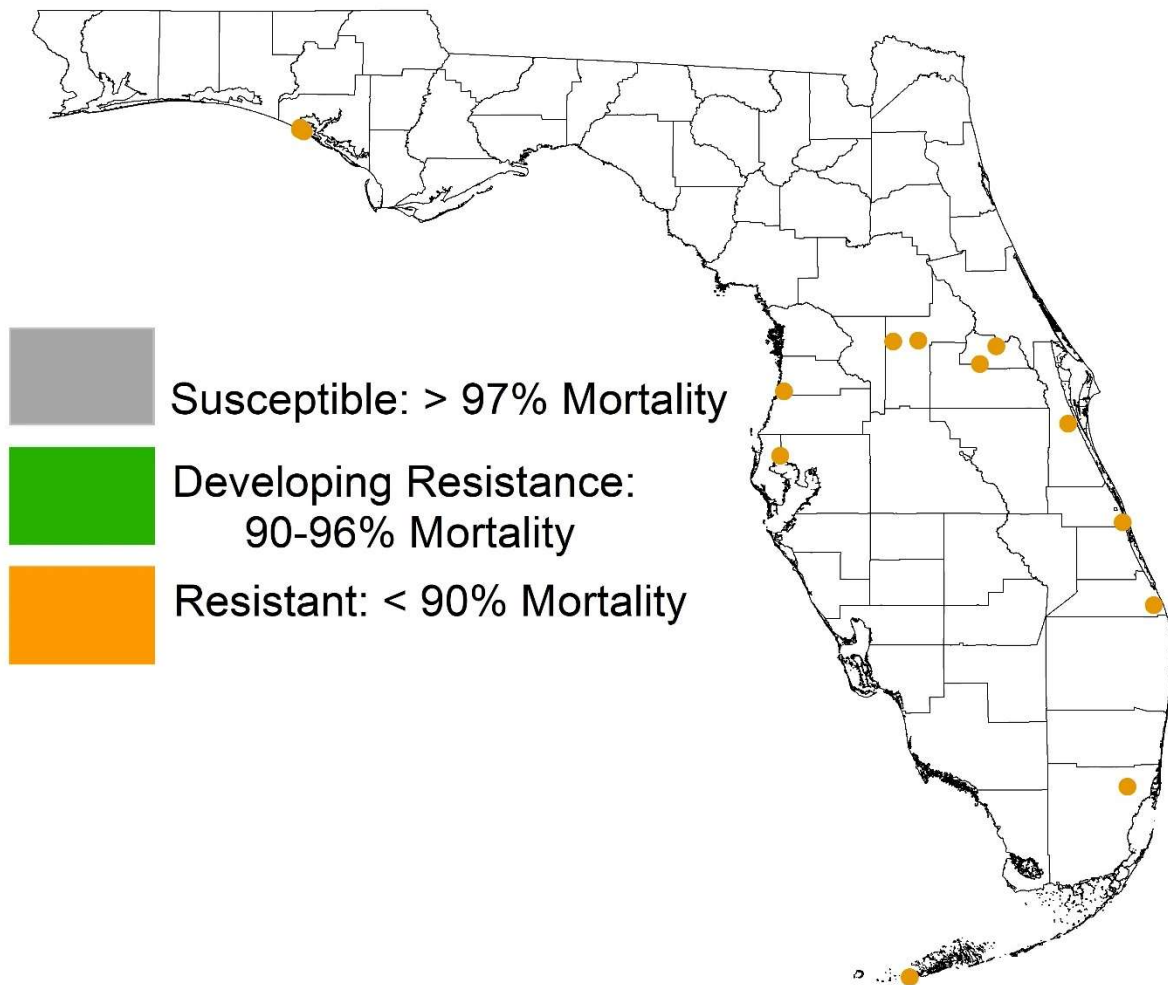
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Species: *Aedes albopictus*
Active Ingredient: sumithrin



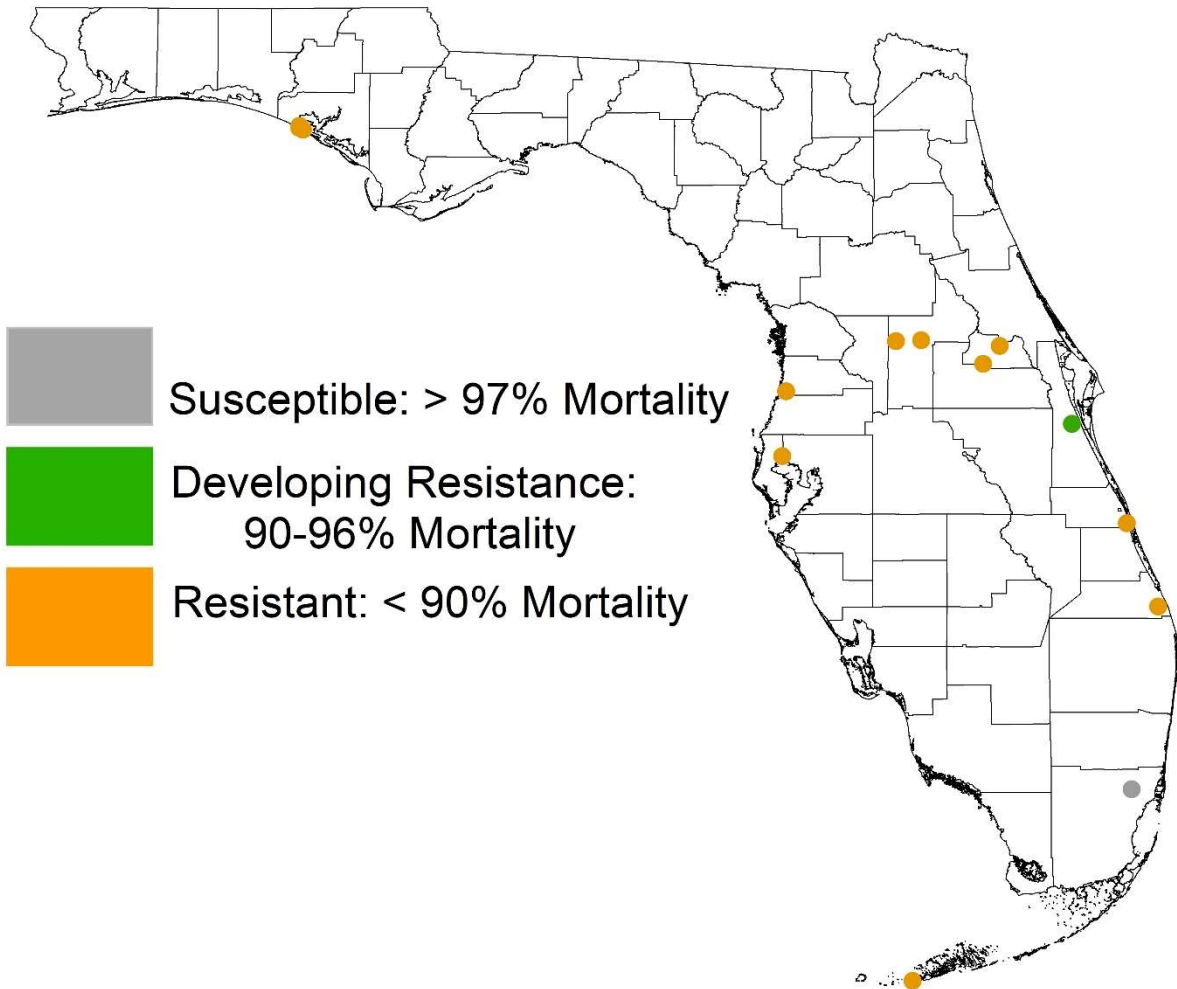
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
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Species: *Culex quinquefasciatus*
Active Ingredient: permethrin



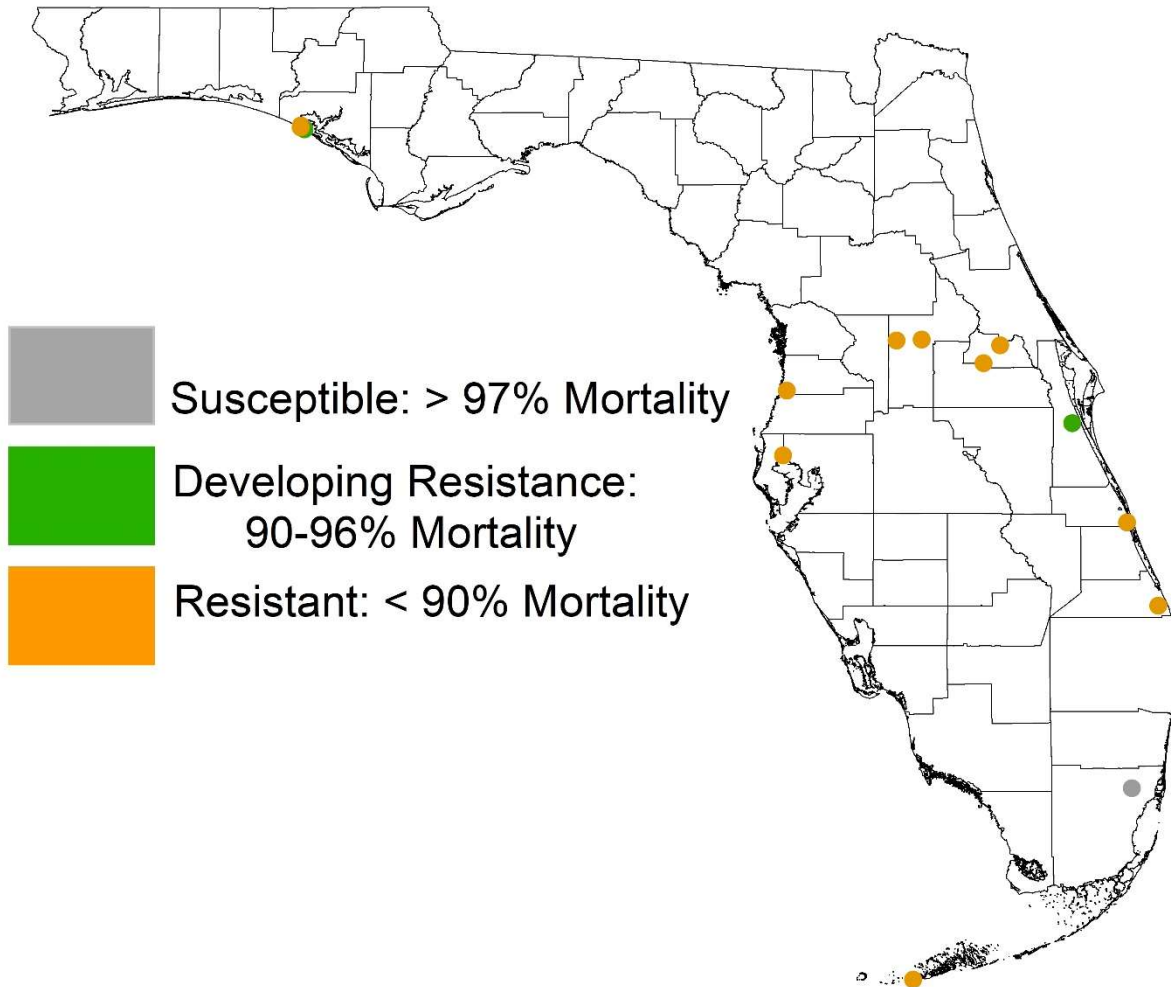
Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
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Species: *Culex quinquefasciatus*
Active Ingredient: malathion



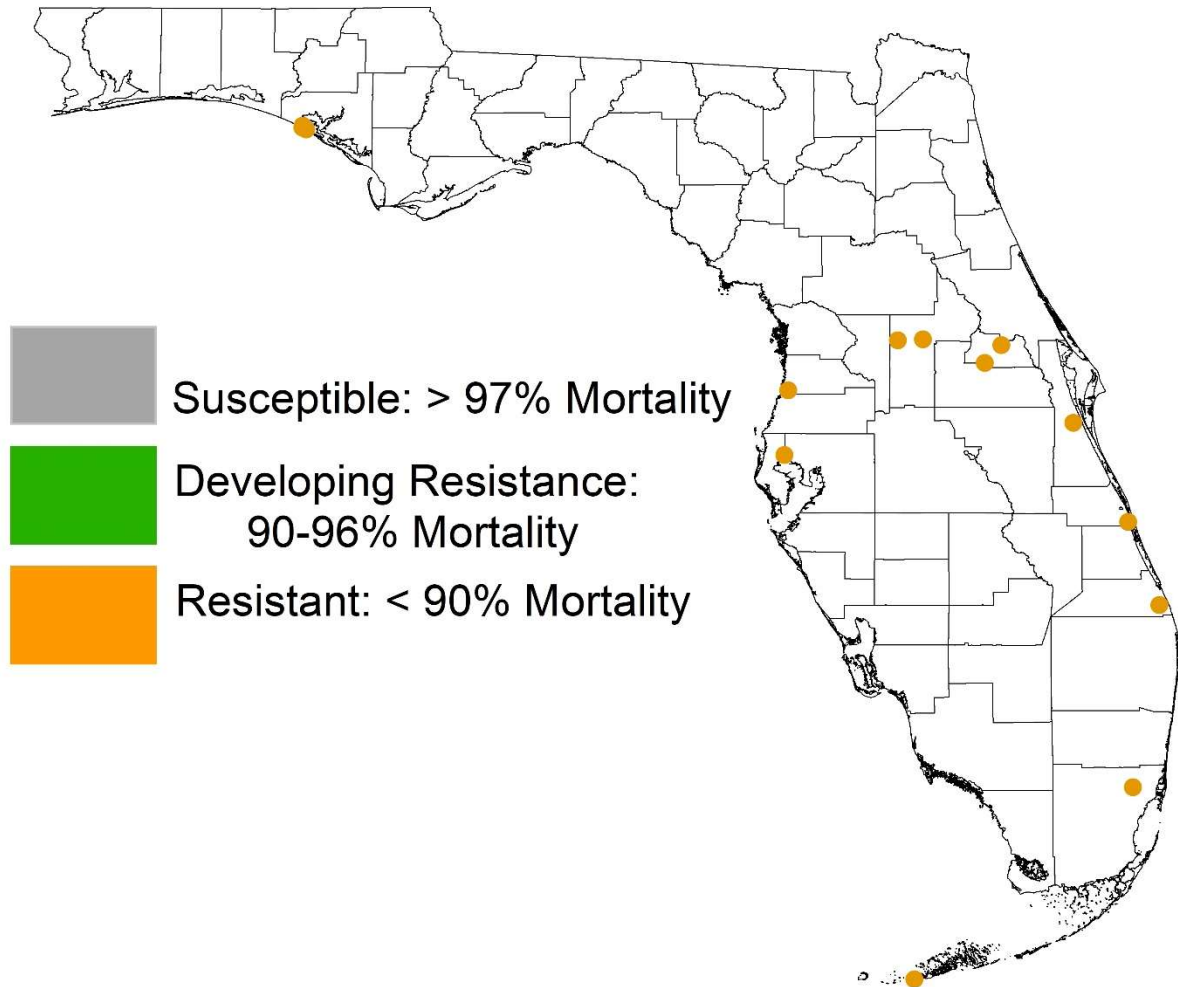
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Species: *Culex quinquefasciatus*
Active Ingredient: naled



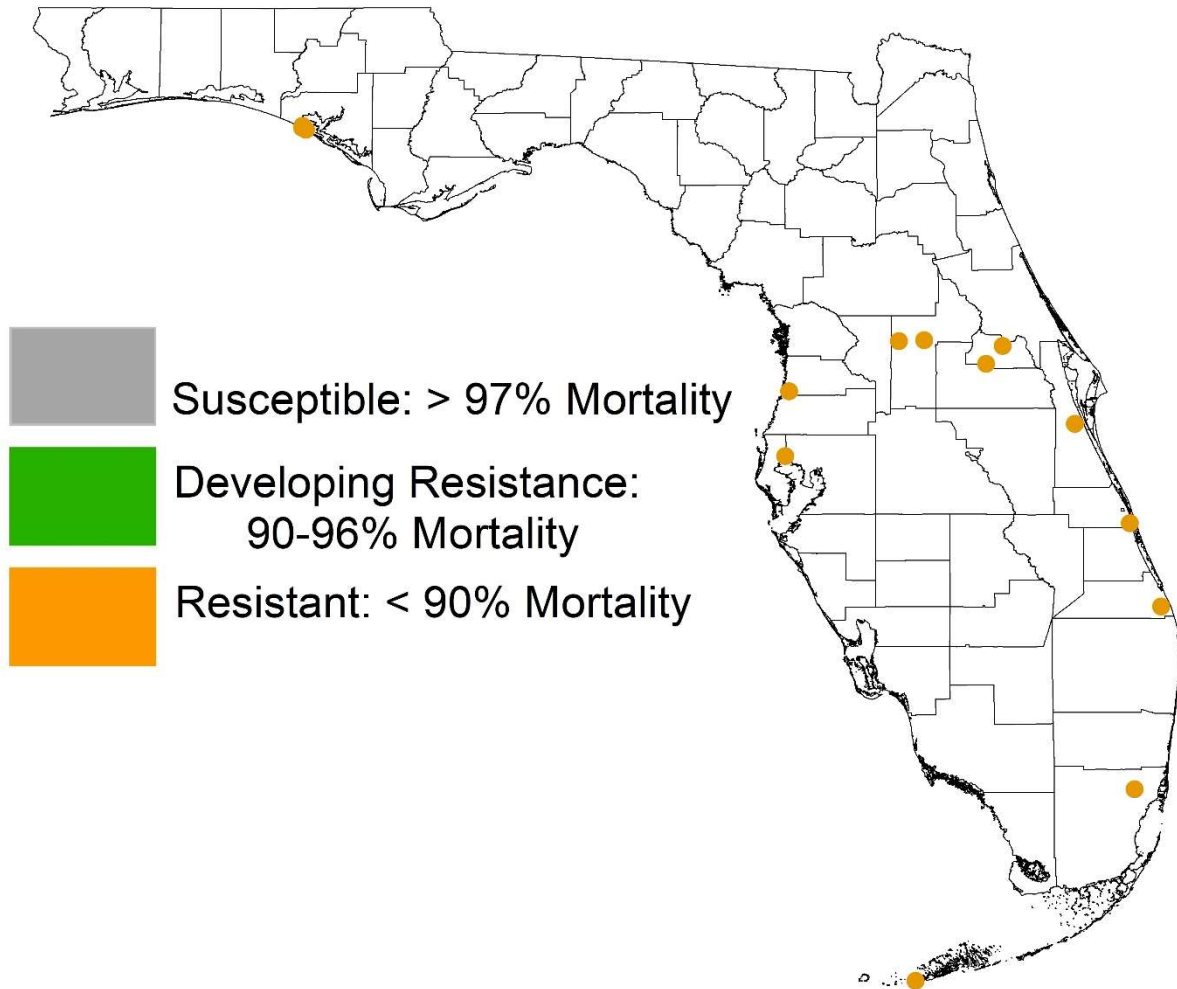
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Species: *Culex quinquefasciatus*
Active Ingredient: deltamethrin



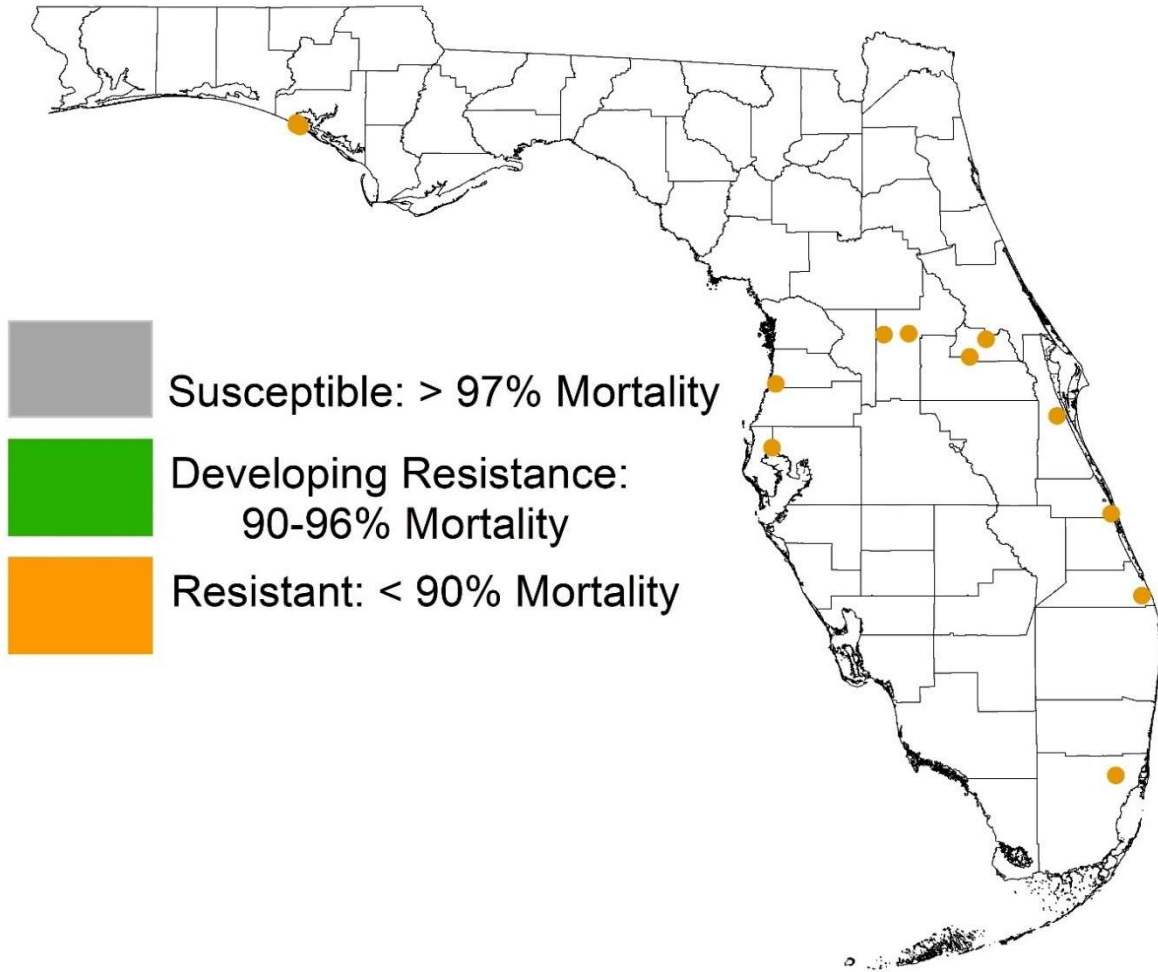
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Species: *Culex quinquefasciatus*
Active Ingredient: etofenprox



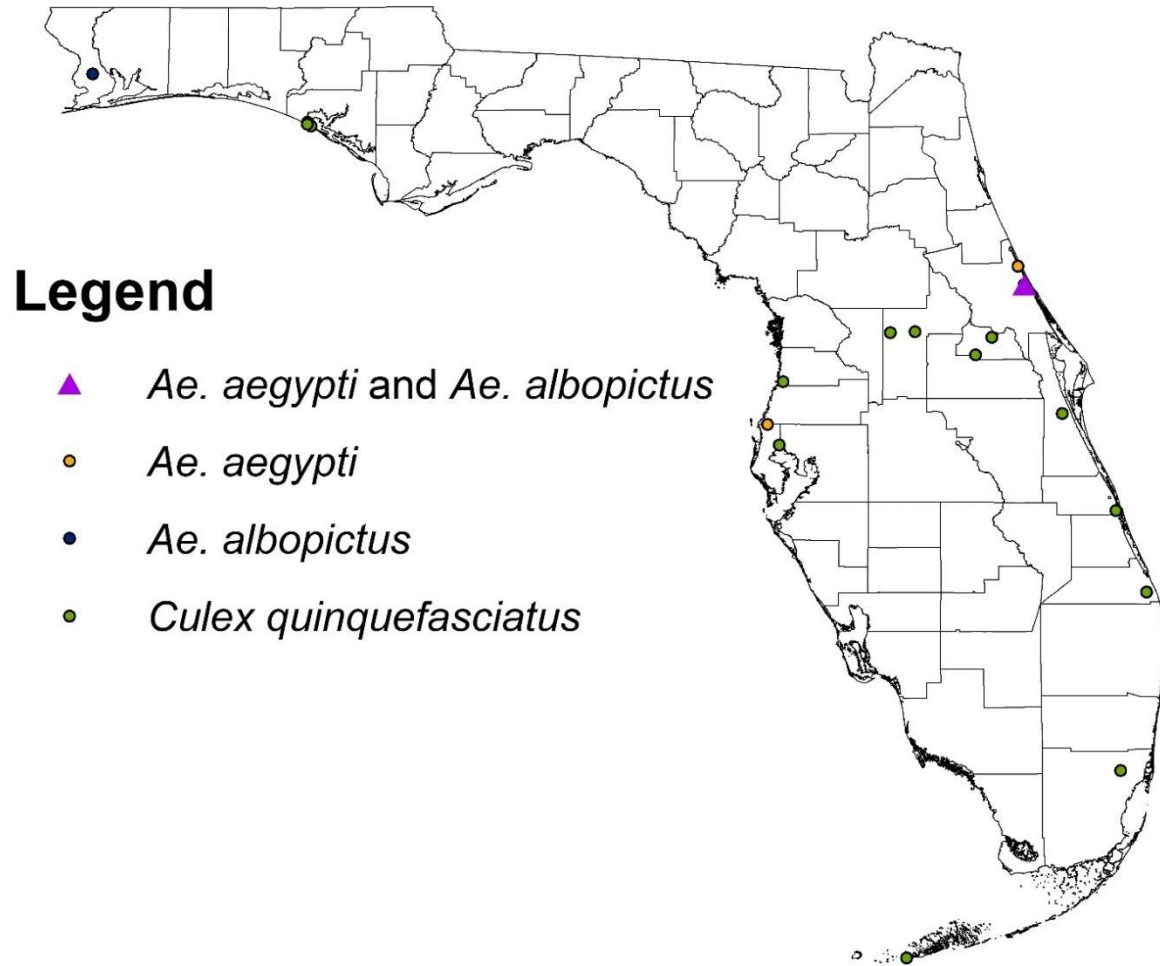
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Species: *Culex quinquefasciatus*
Active Ingredient: sumithrin



Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Locations of domestic mosquito populations tested for insecticide resistance



Credit: E. Buckner, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated October 2019

Assay: CDC bottle bioassay
 Conducted by: UF/IFAS/FMEL – Buckner Lab
 Species: *Aedes aegypti*
 Source: Reared from eggs
 Site: Pinellas County (Sawgrass)
 Address: 1100 Sawgrass Dr., Tarpon Springs, FL 34689
 Coordinates: 28.172633, -82.735169

Date of assay: 7/16/19 – 7/17/19

Following the CDC Guidelines for insecticide resistance monitoring <http://www.cdc.gov/zika/vector/insecticide-resistance.html>, resistance is determined by the percentage of mosquitoes that die (mortality rate) in the diagnostic time.

The data shown below provides:

- Column 1: CDC recommended diagnostic dose (per bottle)
- Column 2: Active ingredient tested
- Column 3: Diagnostic time from FMEL assays; 100% mortality expected at given time using ORL strain of susceptible *Aedes aegypti*;
- Column 4: Site specific *Aedes aegypti*; % mortality at the CDC diagnostic time

1	2	3	4
CDC diagnostic dose (per bottle)	Active ingredient tested	Diagnostic time; 100% mortality expected at given time using ORL strain of susceptible <i>Aedes aegypti</i>	Sawgrass <i>Aedes aegypti</i> ; % mortality at the CDC diagnostic time
43 ug/bottle	Permethrin	15 min	7%
400 ug/bottle	Malathion	30 min	100%
2.25 ug/bottle	Naled	30 min	67%
12.5 ug/bottle	Etofenprox	30 min	3%
20 ug/bottle	Sumethrin	30 min	12%
0.75 ug/bottle	Deltamethrin	15 min	57%

Using the CDC guidelines (<http://www.cdc.gov/zika/vector/insecticide-resistance.html>) on interpreting the data for management purposes:

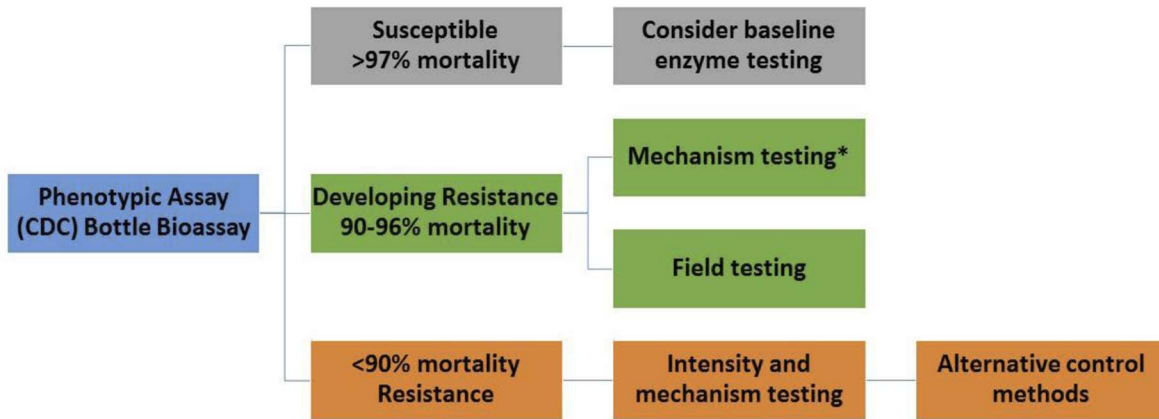
The Sawgrass population of *Aedes aegypti* is resistant to Permethrin, Naled, Etofenprox, Sumethrin and Deltamethrin; Susceptible to Malathion.

Buckner Lab
 UF/IFAS FMEL
 200 9th Street SE
 Vero Beach, FL 32962
 772-226-6606
eva.buckner@ufl.edu



Overview of Insecticide Resistance Testing Algorithm

From: <http://www.cdc.gov/zika/pdfs/guidelines-for-aedes-surveillance-and-insecticide-resistance-testing.pdf>



*Mechanism testing options: enzymes, molecular assays, bottle bioassay with inhibitors