

Quarter 5 (4/01/2020 - 6/30/20) 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

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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://fmel.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 a minimum of the three 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 *Aedes* eggs and *Culex quinquefasciatus* rafts were collected from the traps. Of the 1,724 *Aedes* eggs collected, there was \approx a 64% hatch rate, resulting in 1,109 *Aedes* adults emerging. The 31 *Culex quinquefasciatus* rafts collected had a \approx 82% hatch rate, resulting in 3,546 *Culex quinquefasciatus* adults emerging. The species identification for the resulting *Aedes* adult mosquitoes was *Aedes albopictus*. The *Aedes* eggs processed this quarter came (county followed by site name):
 - a. Indian River
 - i. VLE

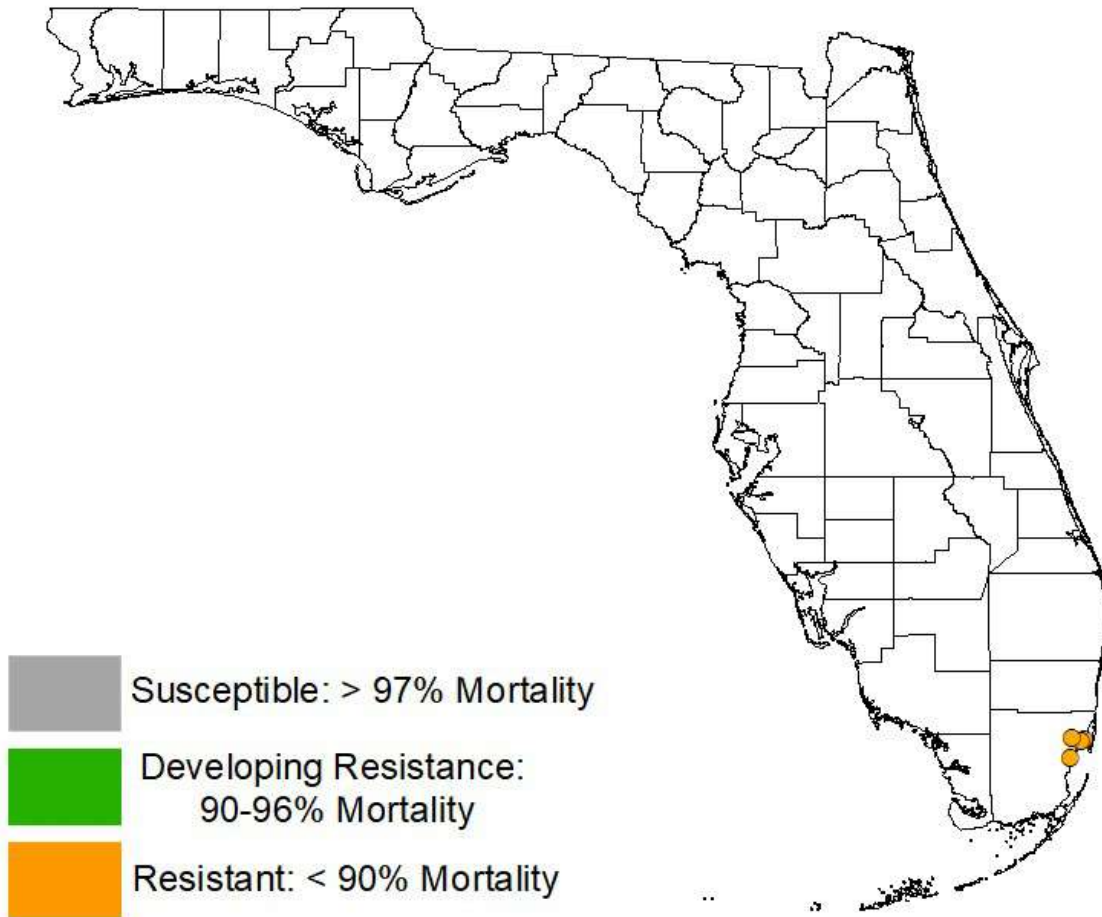
The *Culex quinquefasciatus* rafts processed this quarter came from (county followed by site name):

- b. Citrus
 - i. Rose
 - c. Palm Beach
 - i. Clint Moore
 - ii. Delaware
 - iii. N. Main St.
 - iv. Sysco
 - d. Polk
 - i. Paint Shop
 - e. St. Lucie
 - i. Fairwinds
2. Since the end of the previous quarter, 4 populations of *Aedes aegypti*, 2 populations of *Ae. albopictus* and 10 populations of *Culex quinquefasciatus*, respectively, have been tested 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 - 20 of this report.
3. CDC bottle bioassay testing was performed on mosquitoes from 13 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 be found at <https://fme1.ifas.ufl.edu/>.

Species Tested	County	Site	# of AIs Tested
<i>Aedes aegypti</i>	Miami-Dade	Brickell	4
<i>Aedes aegypti</i>	Miami-Dade	Little Havana	5
<i>Aedes aegypti</i>	Miami-Dade	Flagler Cemetery	5
<i>Aedes aegypti</i>	Miami-Dade	Kings Bay	5
<i>Aedes albopictus</i>	Seminole	Halsey	4
<i>Aedes albopictus</i>	South Walton	Golf Club	4
<i>Culex quinquefasciatus</i>	Citrus	Rose	4
<i>Culex quinquefasciatus</i>	Indian River	Vista Royal	4
<i>Culex quinquefasciatus</i>	Indian River	WB Storm Drain	3
<i>Culex quinquefasciatus</i>	Palm Beach	Clint Moore	3
<i>Culex quinquefasciatus</i>	Palm Beach	Delaware	3
<i>Culex quinquefasciatus</i>	Palm Beach	N. Main St.	1
<i>Culex quinquefasciatus</i>	Palm Beach	Sysco	5
<i>Culex quinquefasciatus</i>	Polk	Paint Shop	2
<i>Culex quinquefasciatus</i>	Seminole	Halsey	2
<i>Culex quinquefasciatus</i>	Seminole	Lake Mary	1

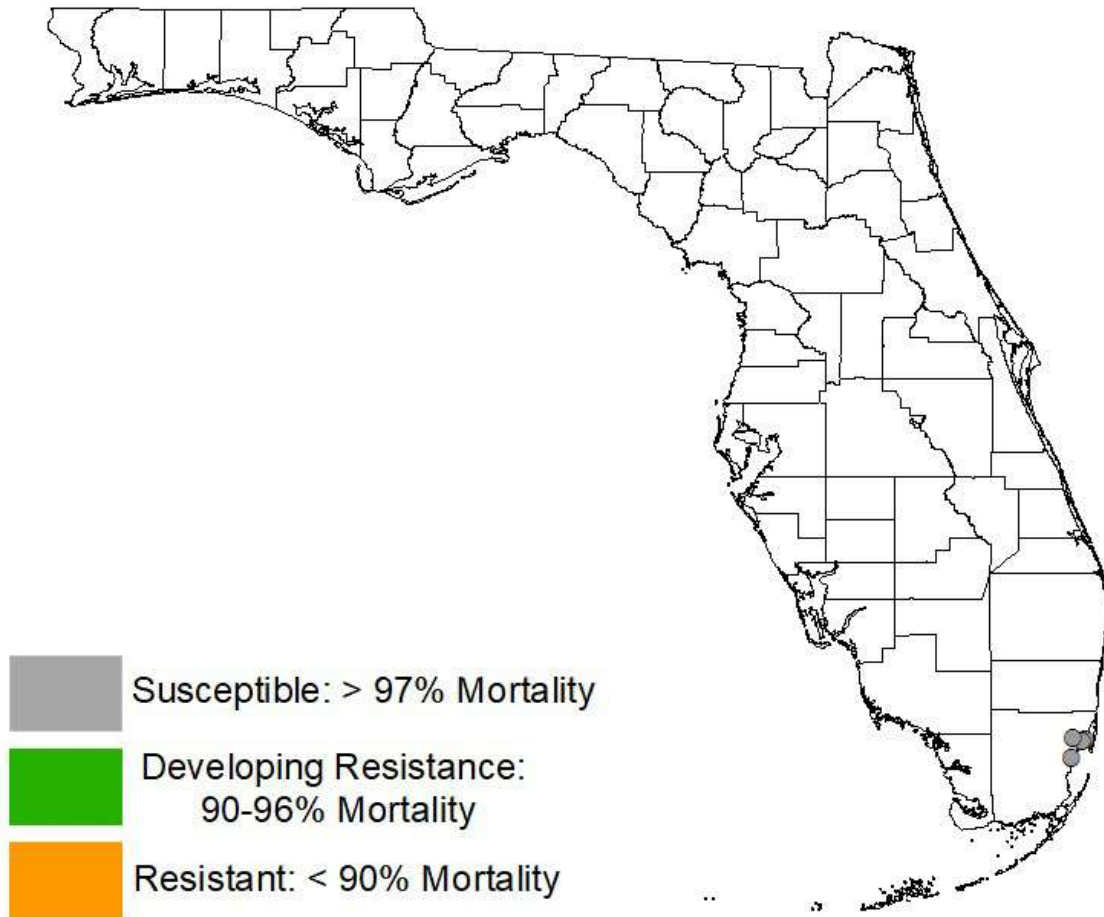
4. The distribution of where *Aedes* eggs and *Culex* egg rafts or larvae were collected from and used in CDC bottle bioassays was mapped and can be found on page 21 of this report. The distribution map can also be found at <https://fmel.ifas.ufl.edu/>.
5. Results of the CDC bottle bioassay testing were distributed as reports by email to Florida mosquito control program managers. Specifically, the results sent to program managers for the Clint Moore, Delaware, and Sysco sites in Palm Beach County, Golf Club site in South Walton County, and Rose site in Citrus County were forwarded to the FDOH Contract Manager to meet requirement of documenting the distribution of results for 3 sites. An example report can be found on pages 22-23.
6. This Quarterly Report in combination with the information posted on the Reporting website at <https://fmel.ifas.ufl.edu/> satisfies this task.
7. All necessary information will be provided in the Final Annual Report.

Species: *Aedes aegypti*
Active Ingredient: permethrin



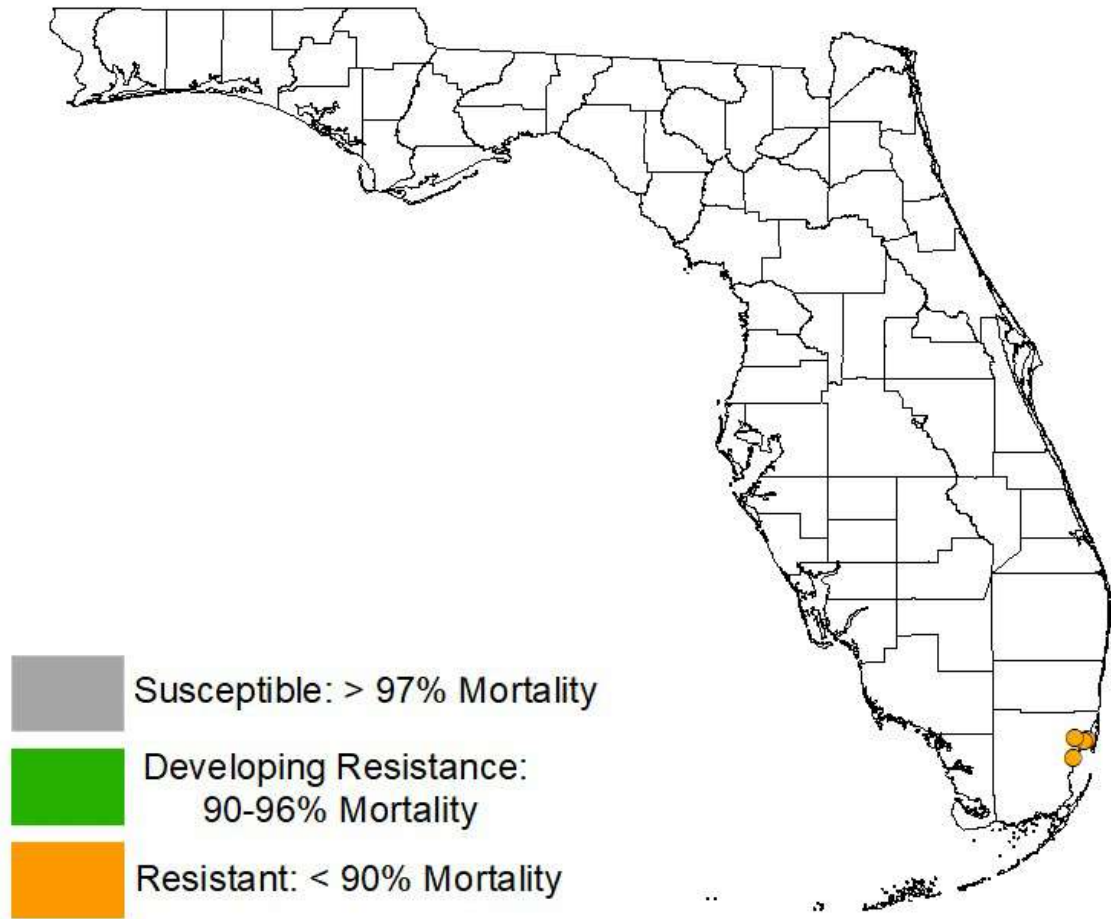
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes aegypti*
Active Ingredient: malathion



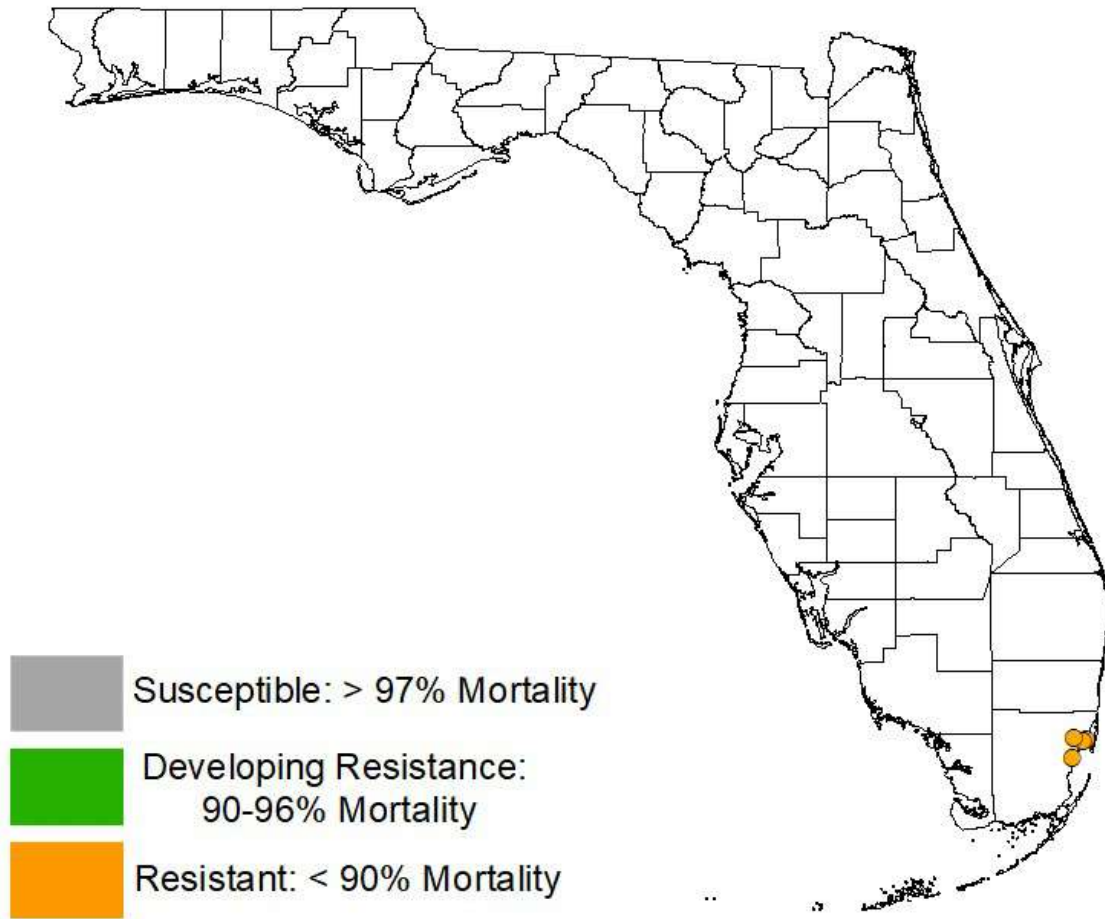
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes aegypti*
Active Ingredient: deltamethrin



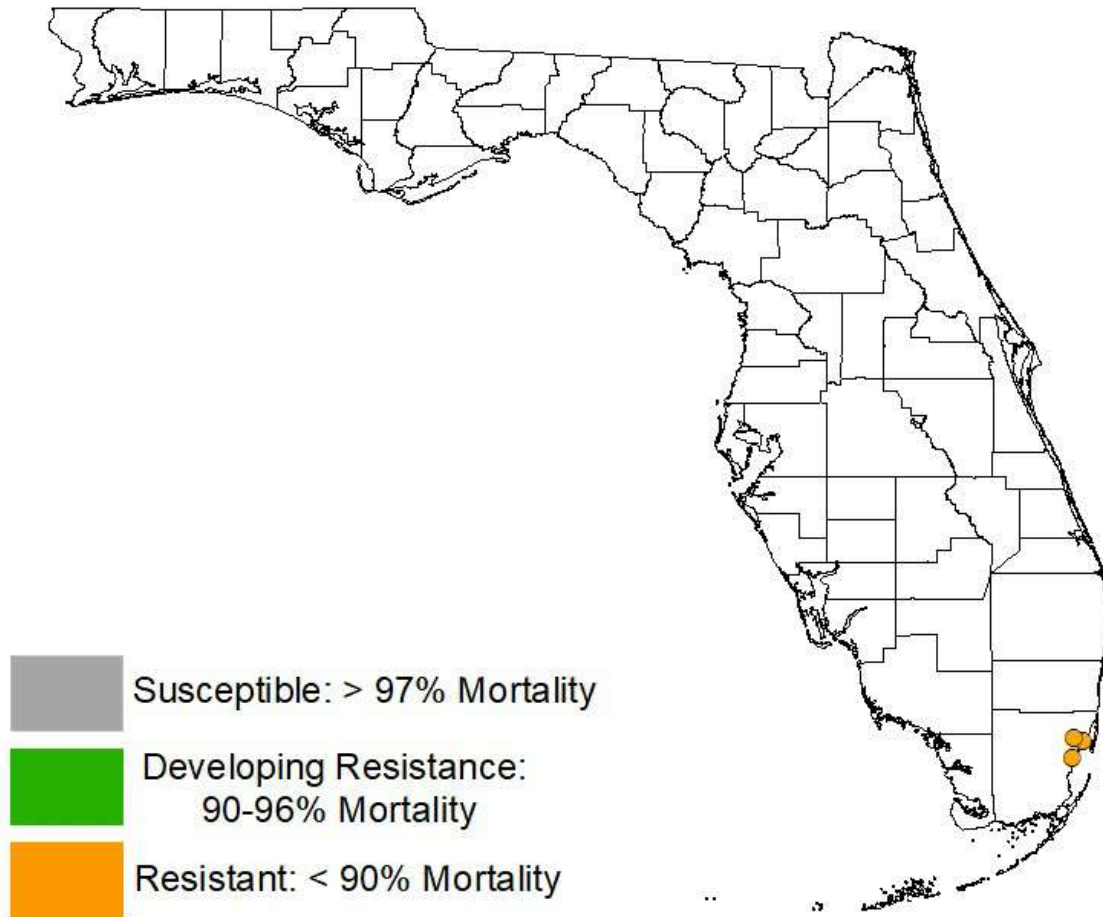
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes aegypti*
Active Ingredient: etofenprox



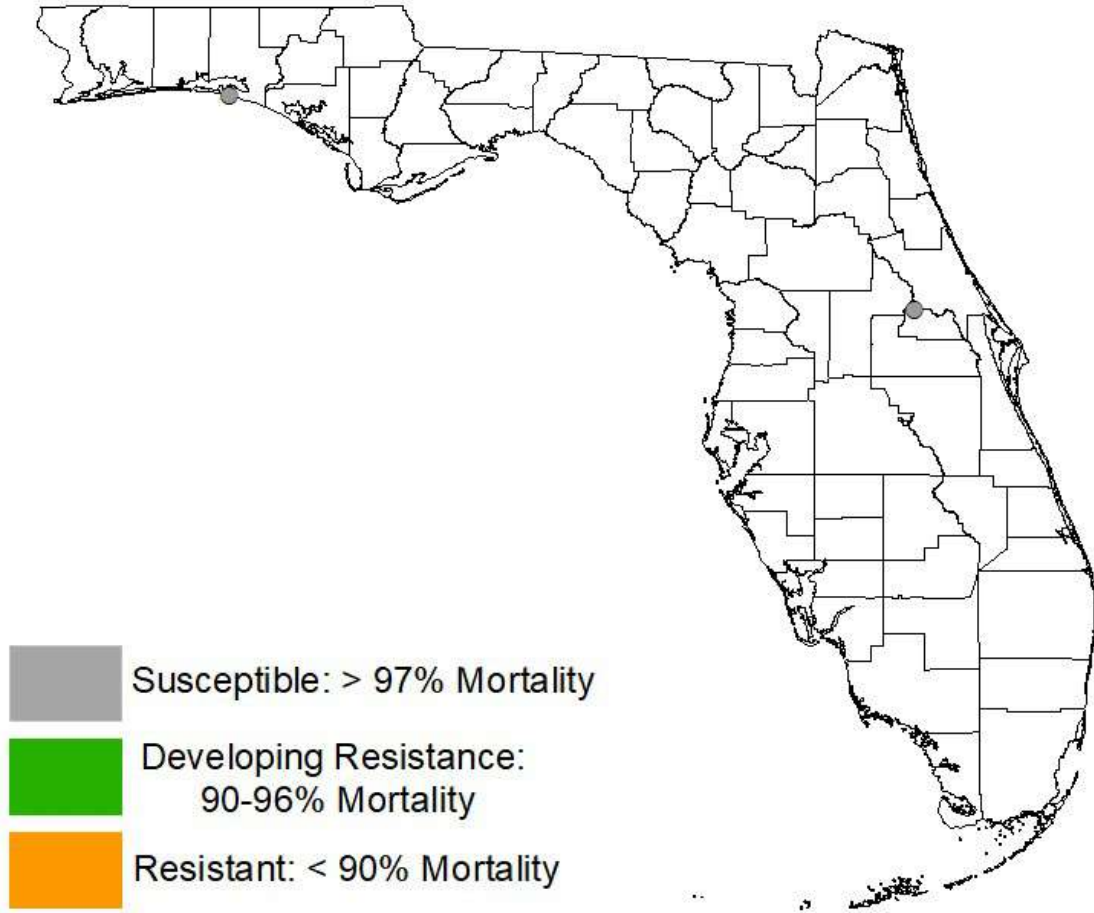
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes aegypti*
Active Ingredient: sumithrin



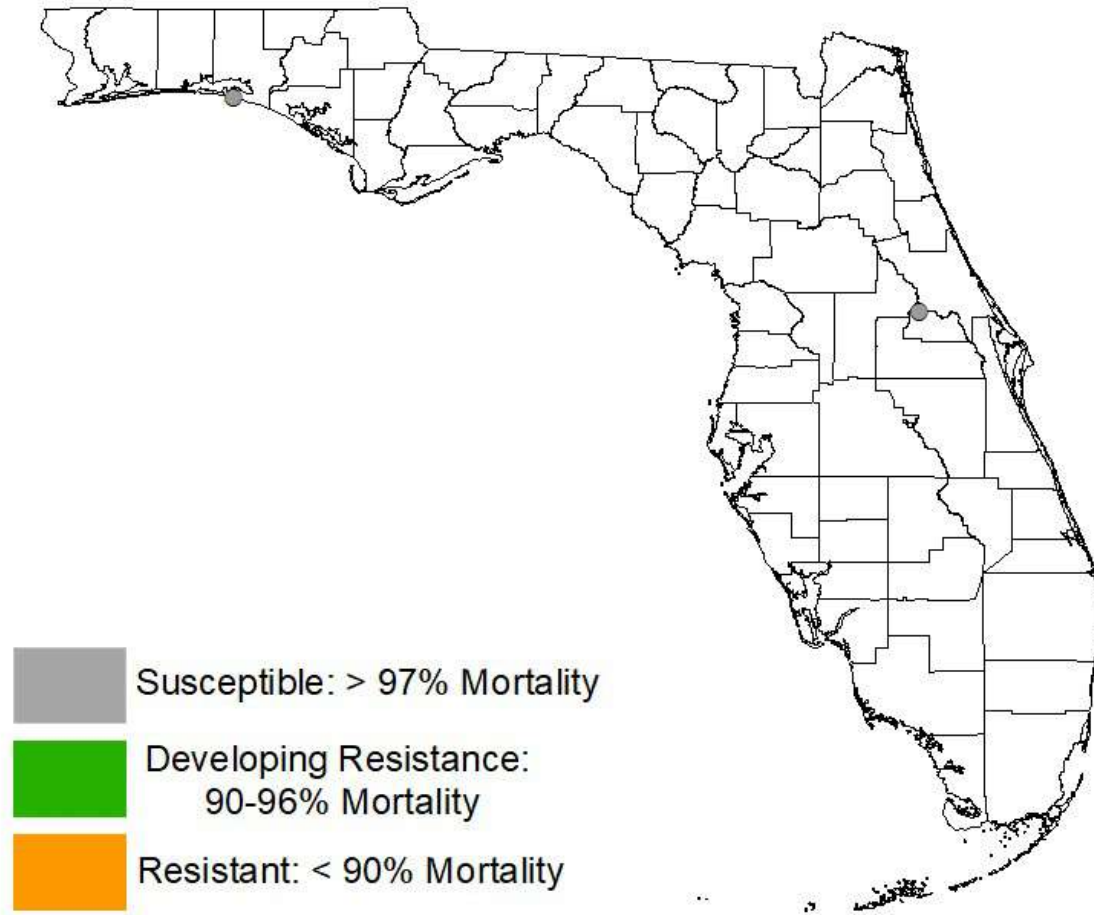
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes albopictus*
Active Ingredient: permethrin



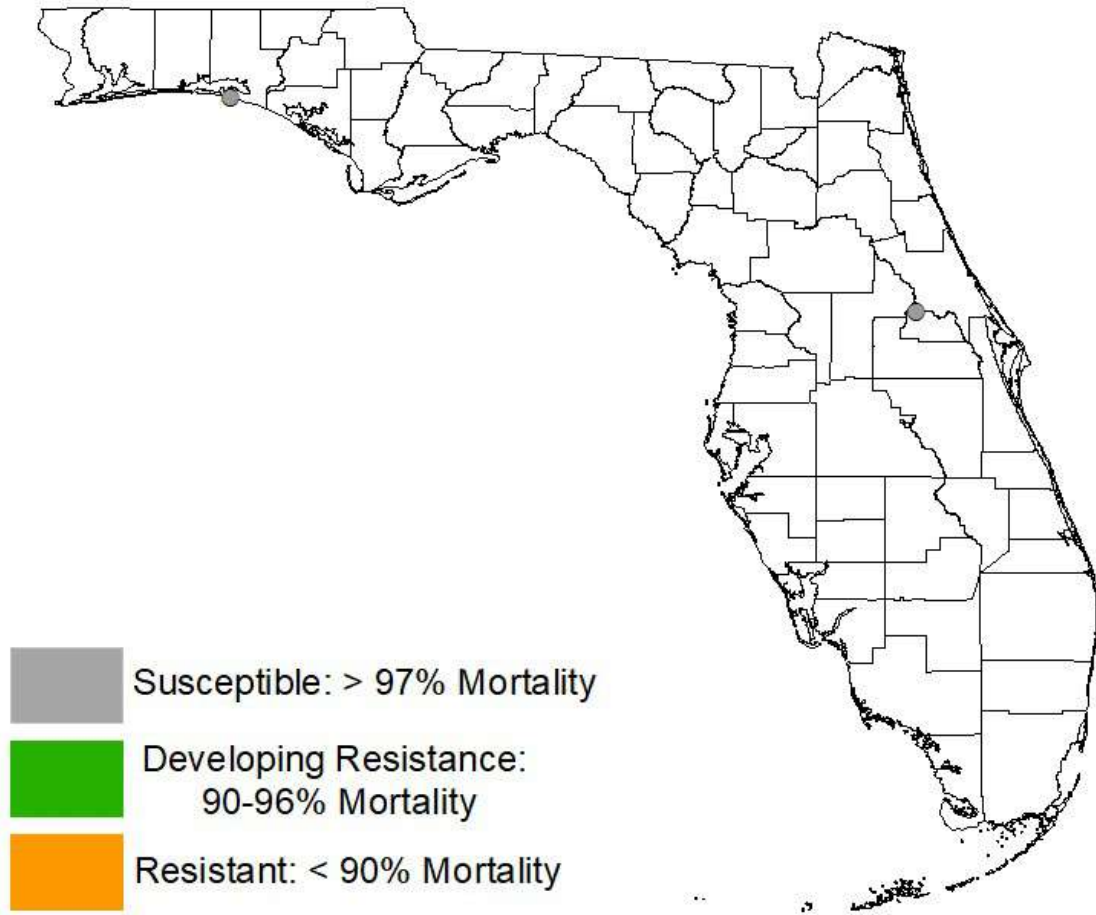
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes albopictus*
Active Ingredient: malathion



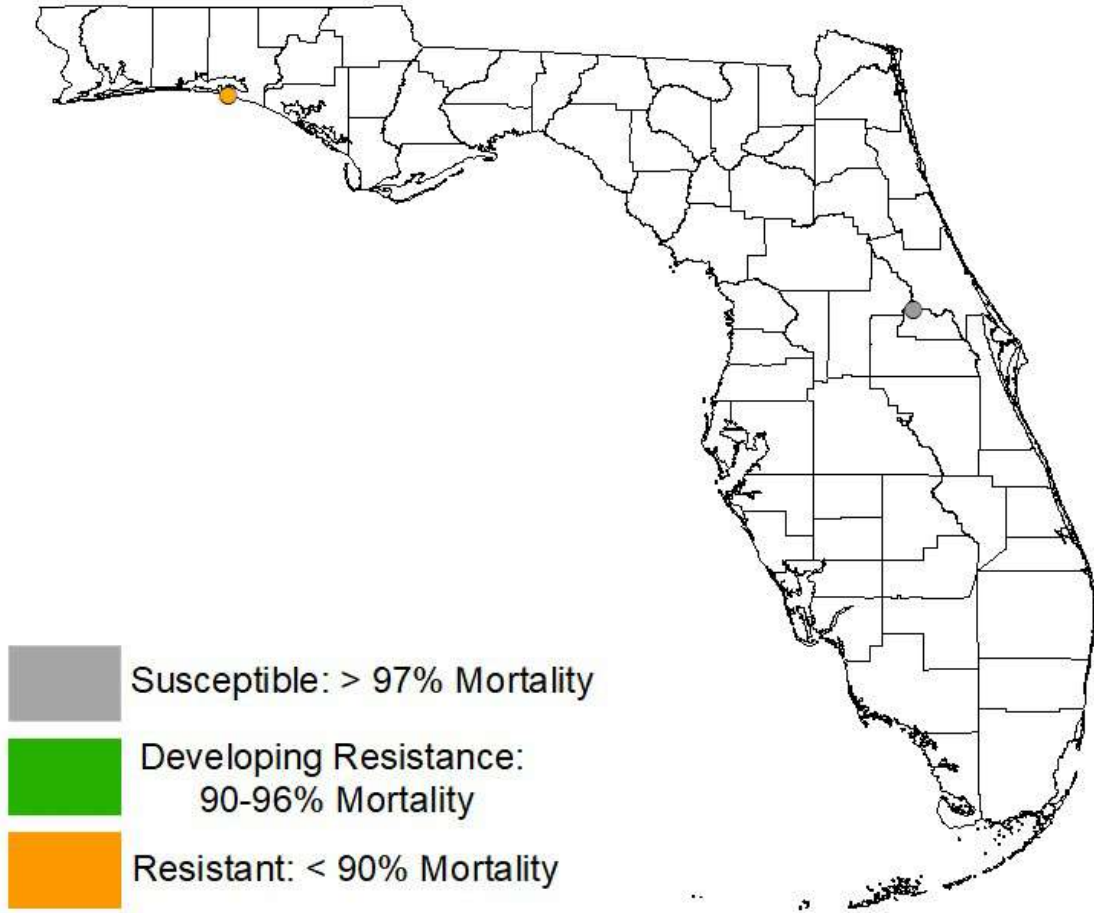
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes albopictus*
Active Ingredient: deltamethrin



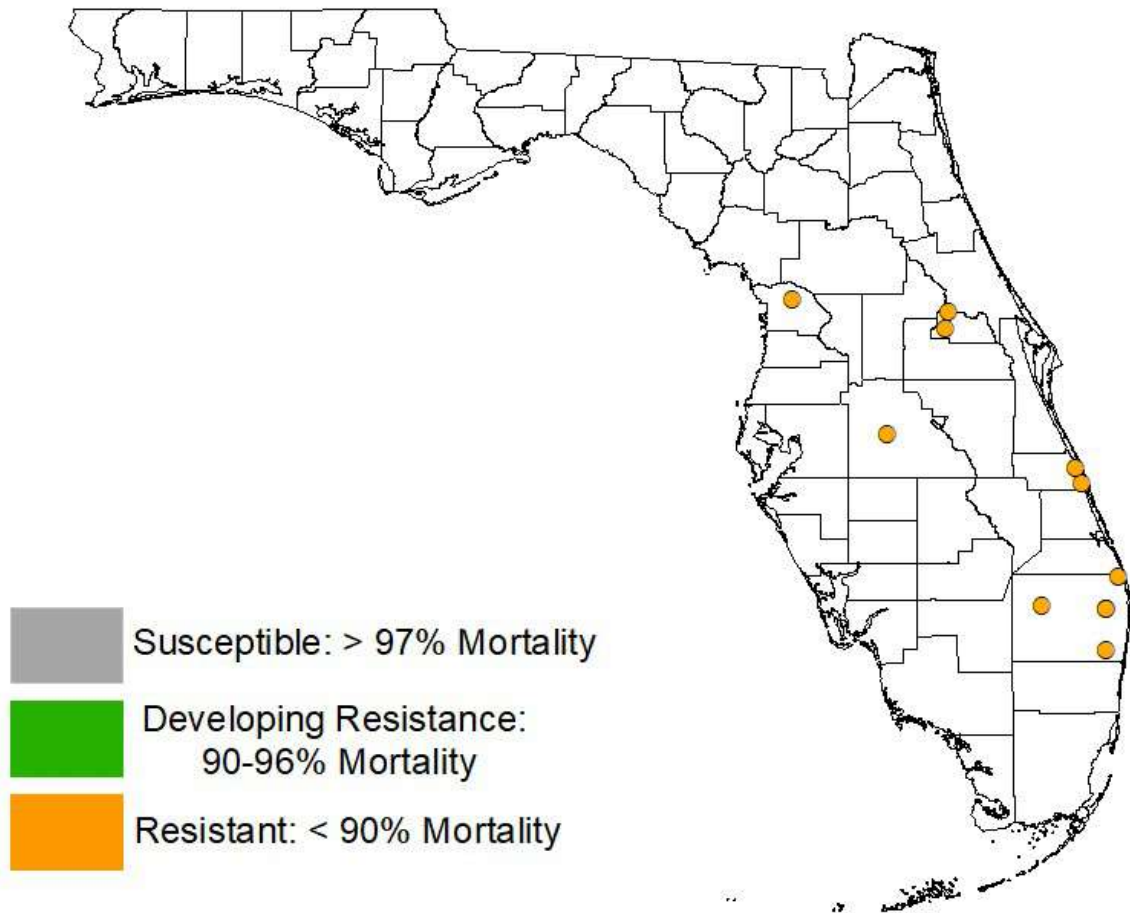
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Aedes albopictus*
Active Ingredient: etofenprox



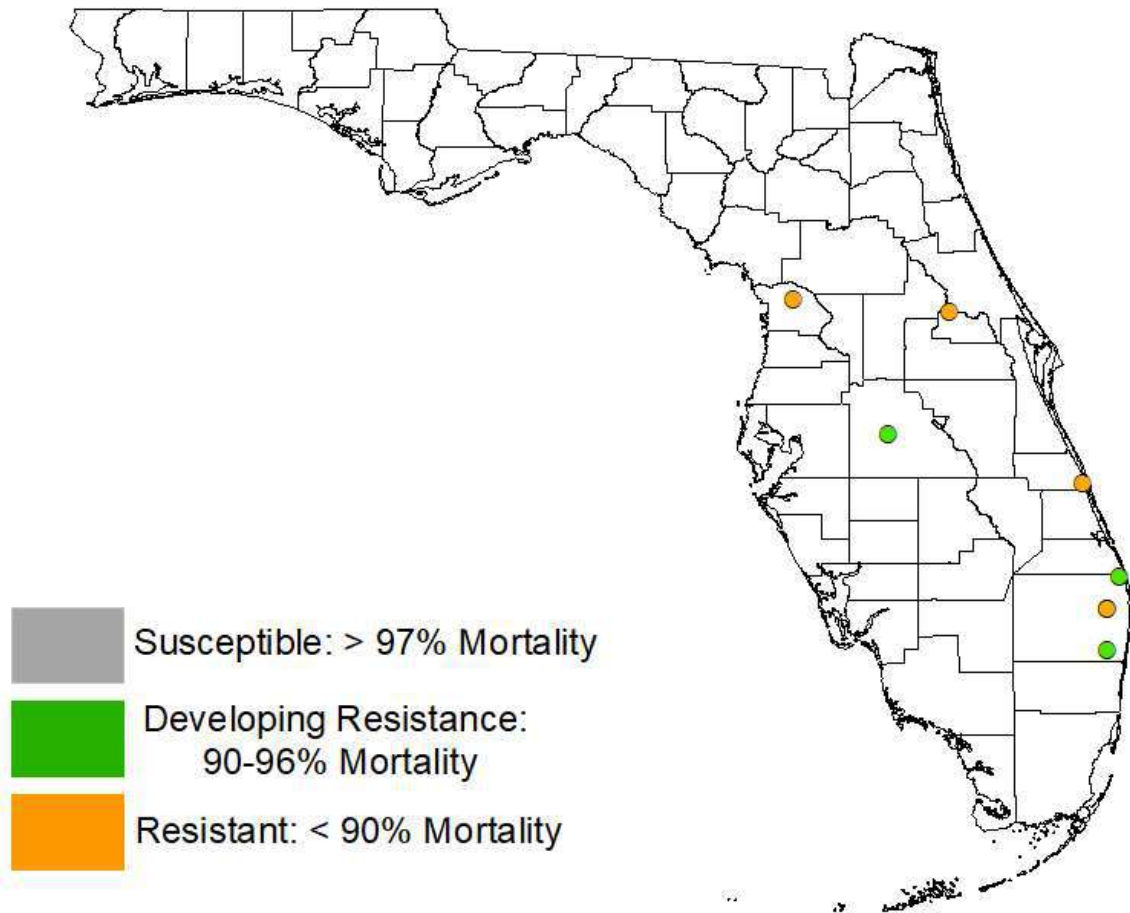
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Culex quinquefasciatus*
Active Ingredient: permethrin



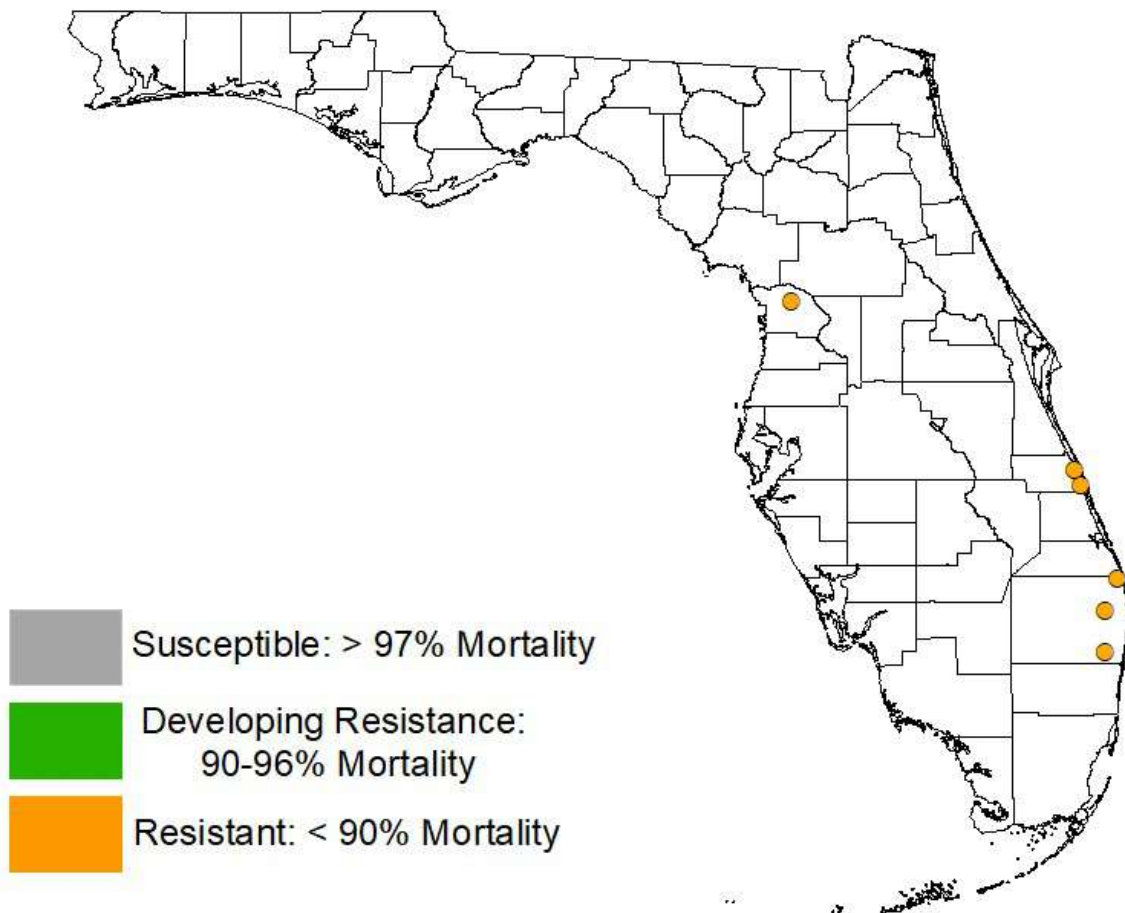
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Culex quinquefasciatus*
Active Ingredient: malathion



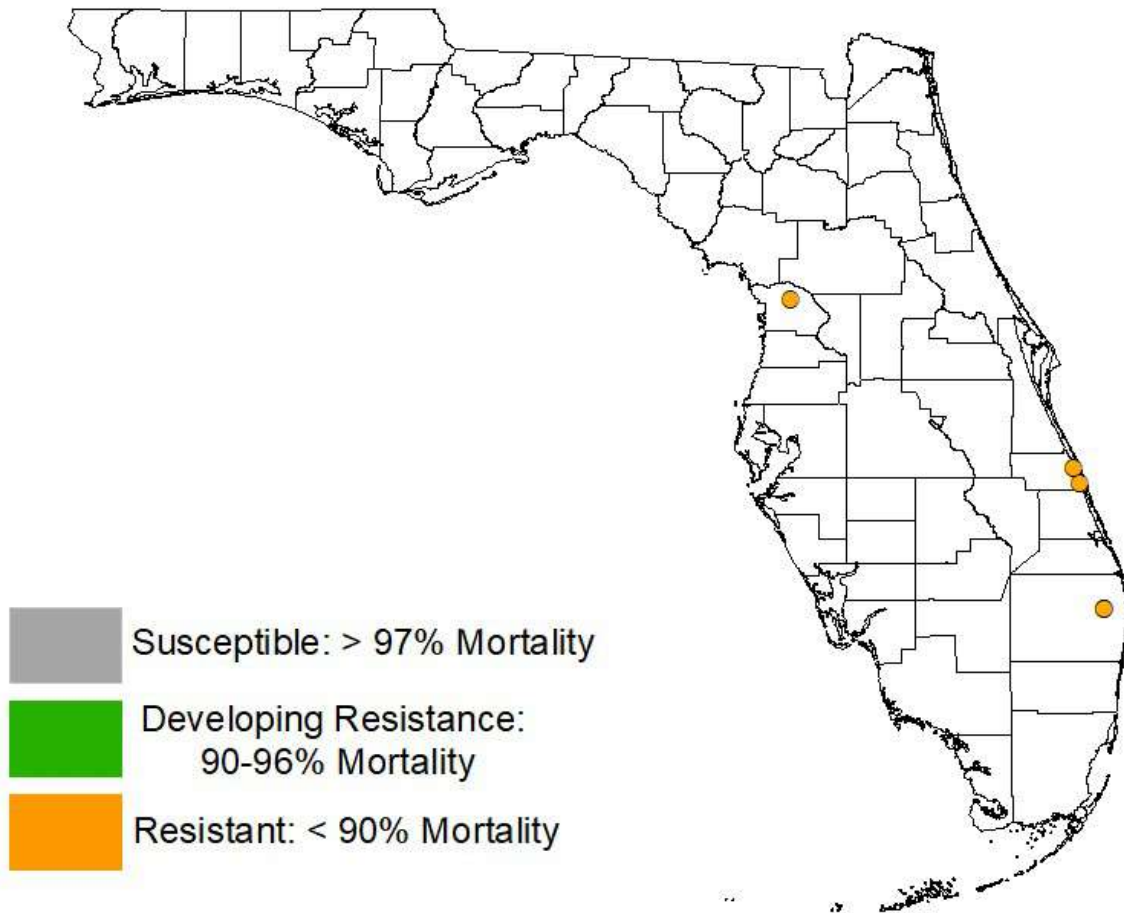
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Culex quinquefasciatus*
Active Ingredient: deltamethrin



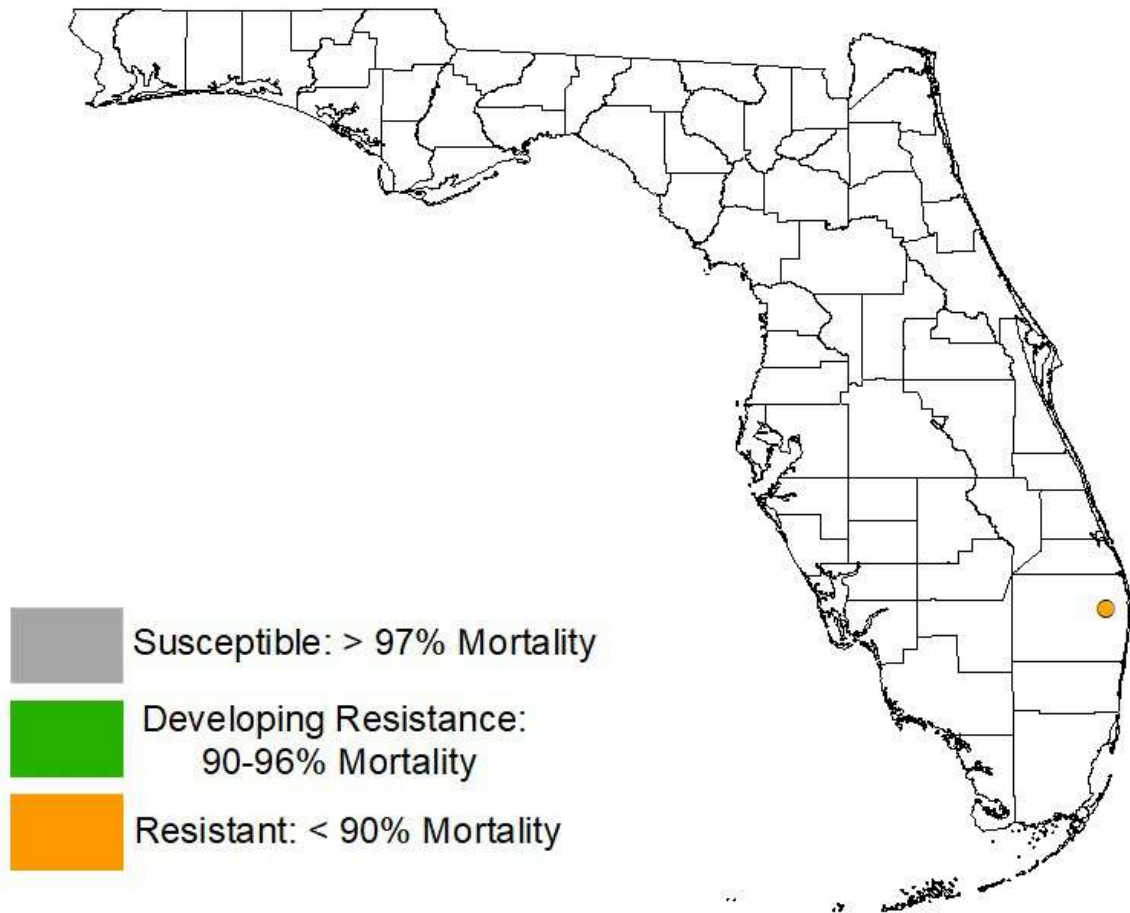
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Culex quinquefasciatus*
Active Ingredient: etofenprox



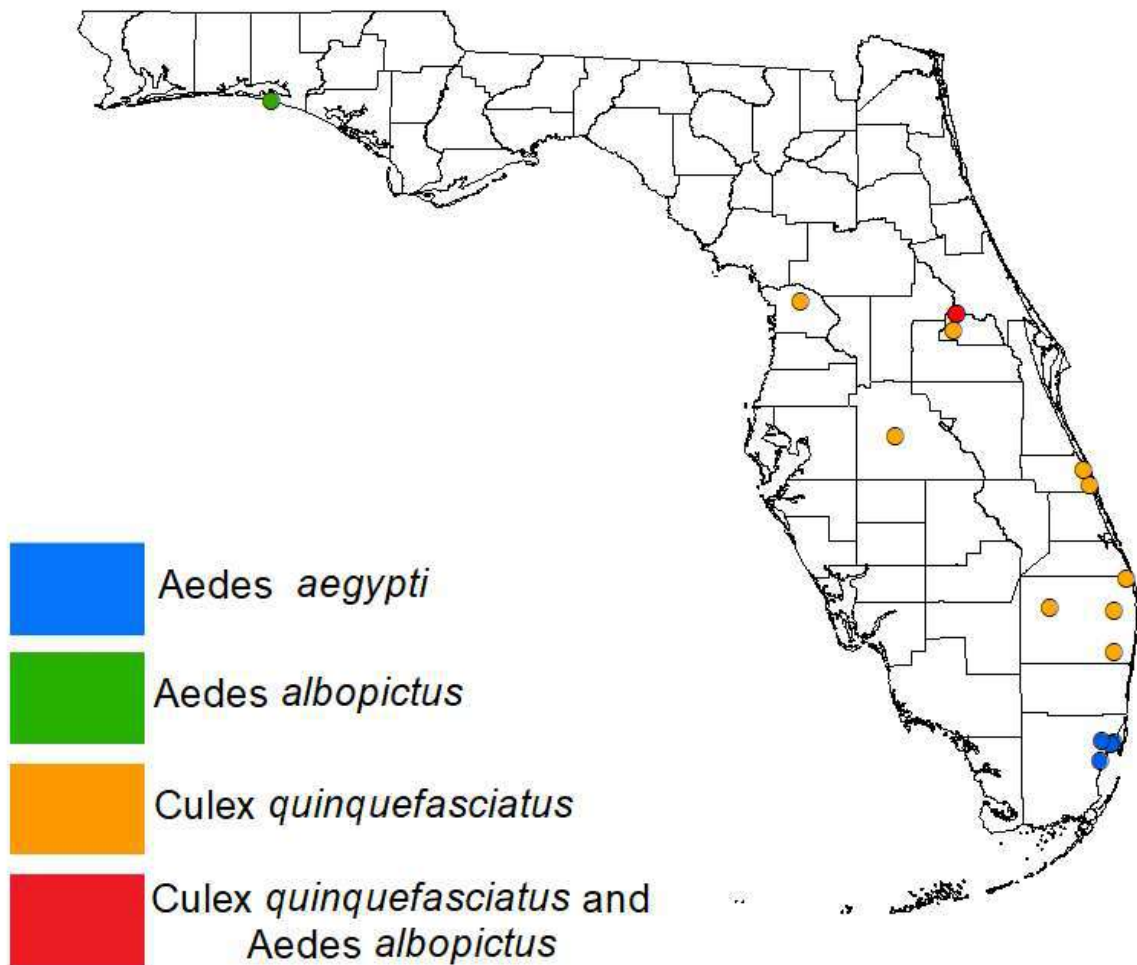
Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Species: *Culex quinquefasciatus*
Active Ingredient: sumithrin



Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Distribution of domestic mosquito populations tested for insecticide resistance April 1 - June 30, 2020



Credit: E. Buckner & D. Ramirez,
UF/IFAS Florida Medical Entomology Laboratory

Assay: CDC bottle bioassay

Conducted by: UF/IFAS FMEL – Buckner Lab

Species: *Aedes albopictus*

Source: Reared from eggs

Site: South Walton County (Golf Club)

Address: Santa Rosa Golf Beach Club, 334 Golf Club Drive, Santa Rosa Beach, FL 32459

Coordinates: 30.355084, -86.240399

Date of assay: 4/2/20

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 a strain of susceptible *Aedes albopictus*

Column 4: Site specific *Aedes albopictus* % mortality at the CDC diagnostic time

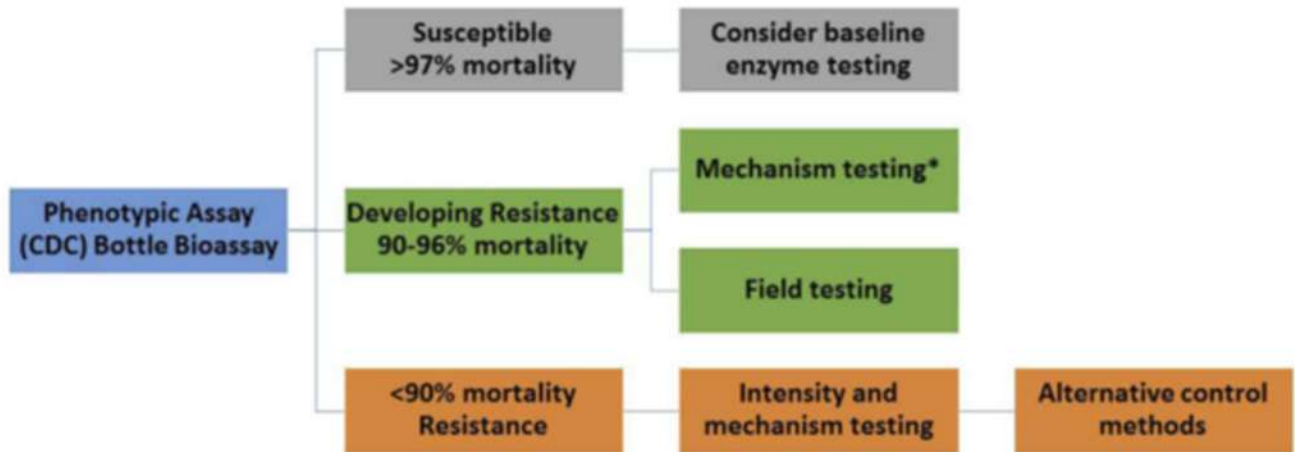
1	2	3	4
CDC diagnostic dose (per bottle)	Active ingredient tested	CDC diagnostic time; 100% mortality expected at given time using strain of susceptible <i>Aedes albopictus</i>	Golf Club <i>Aedes albopictus</i> % mortality at the CDC diagnostic time
43 ug/bottle	Permethrin	10 min	100%
400 ug/bottle	Malathion	30 min	100%
12.5 ug/bottle	Etofenprox	30 min	80%
0.75 ug/ bottle	Deltamethrin	15 min	100%

Using the CDC guidelines (<http://www.cdc.gov/zika/vector/insecticide-resistance.html>) on interpreting the data for management purposes (see page 2 of this document for reference):

The Golf Club population of *Aedes albopictus* is resistant to Etofenprox; susceptible to Permethrin, Malathion and Deltamethrin.

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