Quarter 3 (10/1/2019 – 12/31/2019) 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

13 January 2020
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 a minimum of three 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 20,497 Aedes eggs and 47 Culex quinquefasciatus rafts were collected from the traps. Of the 20,497 Aedes eggs collected, there was ≈ a 51% hatch rate, resulting in 10,392 Aedes adults emerging. The Culex quinquefasciatus larvae collected had a ≈ 79% survivorship rate, resulting in 3,397 Culex quinquefasciatus adults emerging. The species identification for the resulting Aedes adult mosquitoes was Aedes aegypti, Ae. albopictus, and Ae. triseriatus.

The Aedes eggs processed this quarter came from (county followed by site name):

a. Brevard
   i. Satellite High
   ii. Viera
b. Escambia
   i. Extension Office
   ii. Pine Forest Road
   iii. Pond
   iv. Tire Refurb
c. Hernando
   i. CDC-7
   ii. Gupton
   iii. Jackson
d. Miami-Dade
   i. Flagler Cemetery
   ii. Homestead
   iii. Little River
   iv. Miami Beach
   v. Wynwood
e. Monroe
   i. Overseas
f. Pasco
   i. Holiday
   ii. Port Richey
g. Santa Rosa
   i. Rustic Trail
h. South Walton
   i. Golf Club
   ii. County Highway
The *Culex quinquefasciatus* rafts and larvae processed this quarter came from (county followed by site name):

a. Bay  
   i. Cox Grade  

b. Palm Beach  
   i. Sysco  

c. Seminole County  
   i. 2nd Street  
   ii. Center Street  
   iii. Halsey  
   iv. Lake Mary  
   v. Lazy Acres  

2. Since the end of the previous quarter, 3 populations of *Aedes aegypti* and 4 populations of *Ae. albopictus* and 2 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 8 - 25 of this report.

3. CDC bottle bioassay testing was performed on mosquitoes from 9 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://fmel.ifas.ufl.edu/](https://fmel.ifas.ufl.edu/).

<table>
<thead>
<tr>
<th>Species Tested</th>
<th>County</th>
<th>Site</th>
<th># of AIs Tested</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Aedes aegypti</em></td>
<td>Monroe</td>
<td>Overseas</td>
<td>6</td>
</tr>
<tr>
<td><em>Aedes aegypti</em></td>
<td>Pasco</td>
<td>Holiday</td>
<td>6</td>
</tr>
<tr>
<td><em>Aedes aegypti</em></td>
<td>Pasco</td>
<td>Port Richey</td>
<td>3</td>
</tr>
<tr>
<td><em>Aedes albopictus</em></td>
<td>Brevard</td>
<td>Viera</td>
<td>6</td>
</tr>
<tr>
<td><em>Aedes albopictus</em></td>
<td>Hernando</td>
<td>CDC7</td>
<td>3</td>
</tr>
<tr>
<td><em>Aedes albopictus</em></td>
<td>Hernando</td>
<td>Gupton</td>
<td>6</td>
</tr>
<tr>
<td><em>Aedes albopictus</em></td>
<td>Hernando</td>
<td>Jackson</td>
<td>5</td>
</tr>
<tr>
<td><em>Culex quinquefasciatus</em></td>
<td>Bay</td>
<td>Cox Grade</td>
<td>6</td>
</tr>
<tr>
<td><em>Culex quinquefasciatus</em></td>
<td>Seminole</td>
<td>Center Street</td>
<td>6</td>
</tr>
</tbody>
</table>

4. The locations of where *Aedes* and *Culex* eggs or larvae were collected from were mapped and can be found on page 26-27 of this report. The distribution map can also be found at [https://fmel.ifas.ufl.edu/](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 in Bay, Hernando, and Seminole Counties for the
sites Cox Grade, Gupton, and Center Street, respectively, this quarter. An example report can be found on pages 28-29.

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

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes aegypti*
Active Ingredient: malathion

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes aegypti*
Active Ingredient: naled

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes aegypti*
Active Ingredient: deltamethrin

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes aegypti*
Active Ingredient: etofenprox

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes aegypti*
Active Ingredient: permethrin

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes albopictus*
Active Ingredient: permethrin

- **Susceptible:** > 97% Mortality
- **Developing Resistance:** 90-96% Mortality
- **Resistant:** < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes albopictus*
Active Ingredient: malathion

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31, 2019
Species: *Aedes albopictus*
Active Ingredient: naled

- Susceptible: > 97% Mortality
- Developing Resistance: 90-96% Mortality
- Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes albopictus*
Active Ingredient: deltamethrin

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes albopictus*
Active Ingredient: etofenprox

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Aedes albopictus*
Active Ingredient: sumithrin

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Culex quinquefasciatus*
Active Ingredient: permethrin

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Culex quinquefasciatus*
Active Ingredient: malathion

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Culex quinquefasciatus*
Active Ingredient: naled

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory,
University of Florida, IFAS
Updated December 31 2019
Species: *Culex quinquefasciatus*
Active Ingredient: deltamethrin

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Culex quinquefasciatus*
Active Ingredient: etofenprox

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Species: *Culex quinquefasciatus*
Active Ingredient: sumithrin

Susceptible: > 97% Mortality
Developing Resistance: 90-96% Mortality
Resistant: < 90% Mortality

Credit: E. Buckner & D. Ramirez, Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31 2019
Distribution of *Aedes aegypti* and *Ae. albopictus* populations collected October – December 2019

Legend

- Green circle: *Ae. aegypti* and *Ae. albopictus*
- Orange circle: *Ae. aegypti*
- Blue circle: *Ae. albopictus*

Credit: E. Buckner & D. Ramirez Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31, 2019
Distribution of *Culex quinquefasciatus* populations collected October – December 2019

Legend

- *Culex quinquefasciatus*

Credit: E. Buckner & D. Ramirez Florida Medical Entomology Laboratory, University of Florida, IFAS
Updated December 31, 2019
Assay: CDC bottle bioassay  
Conducted by: UF/IFAS/FMEL – Buckner Lab  
Species: Aedes albopictus  
Source: Reared from eggs  
Site: Hernando County (Gupton)  
Address: 16212 Gupton Street, Brooksville, FL, 34613  
Coordinates: 28.54562, -82.46238  

Date of assay: 12/16/19 - 12/17/19

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 using susceptible strain of Aedes albopictus  
Column 4: Site specific Aedes albopictus; % mortality at the FMEL diagnostic time

<table>
<thead>
<tr>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CDC diagnostic dose (per bottle)</td>
<td>Active ingredient tested</td>
<td>FMEL diagnostic time given using strain of susceptible Aedes albopictus</td>
</tr>
<tr>
<td>43 ug/bottle</td>
<td>Permethrin</td>
<td>15 min</td>
<td>96%</td>
</tr>
<tr>
<td>400 ug/bottle</td>
<td>Malathion</td>
<td>30 min</td>
<td>90%</td>
</tr>
<tr>
<td>2.25 ug/bottle</td>
<td>Naled</td>
<td>15 min</td>
<td>100%</td>
</tr>
<tr>
<td>12.5 ug/bottle</td>
<td>Etofenprox</td>
<td>30 min</td>
<td>100%</td>
</tr>
<tr>
<td>0.75 ug/bottle</td>
<td>Deltamethrin</td>
<td>15 min</td>
<td>97%</td>
</tr>
<tr>
<td>20 ug/bottle</td>
<td>Sumithrin</td>
<td>15 min</td>
<td>100%</td>
</tr>
</tbody>
</table>


The Gupton population of Aedes albopictus is developing resistance to Permethrin and Malathion; susceptible to Naled, Etofenprox, Deltamethrin and Sumithrin.

Date: 12/29/19

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


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