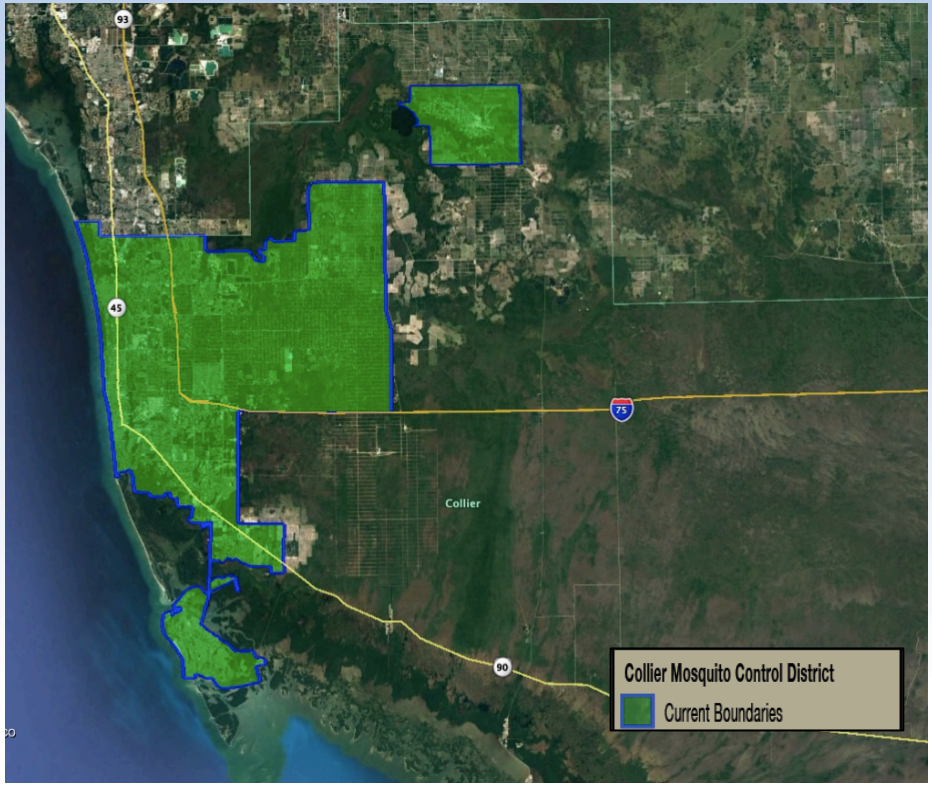
Insecticide resistance status and management in Collier County, Florida

Keira J. Lucas, Rebecca Heinig, Rachel Bales

Deputy Executive Director

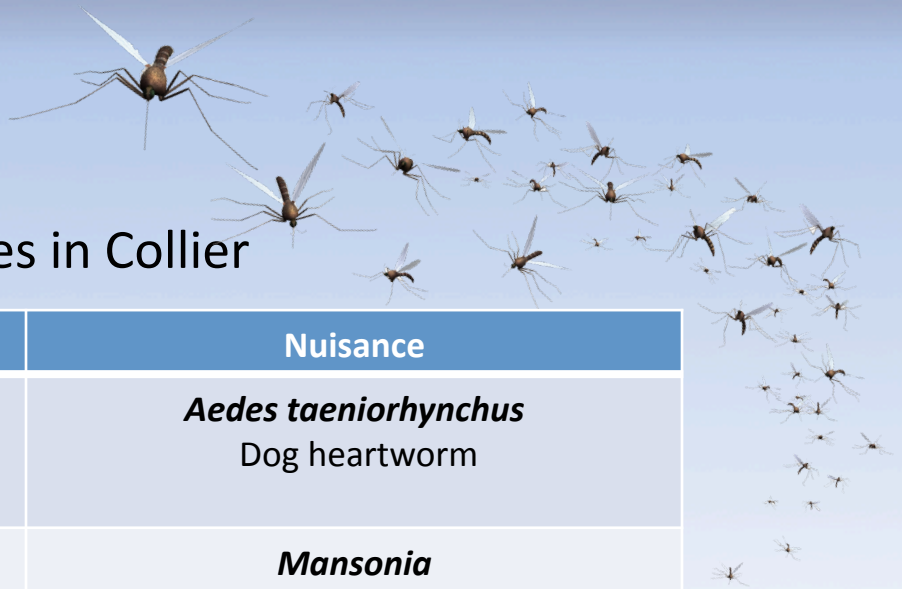
Collier Mosquito Control District

Collier Mosquito Control District



COLLIER MOSQUITO CONTROL DISTRICT

Species of Concern



More than 50 species of mosquitoes in Collier

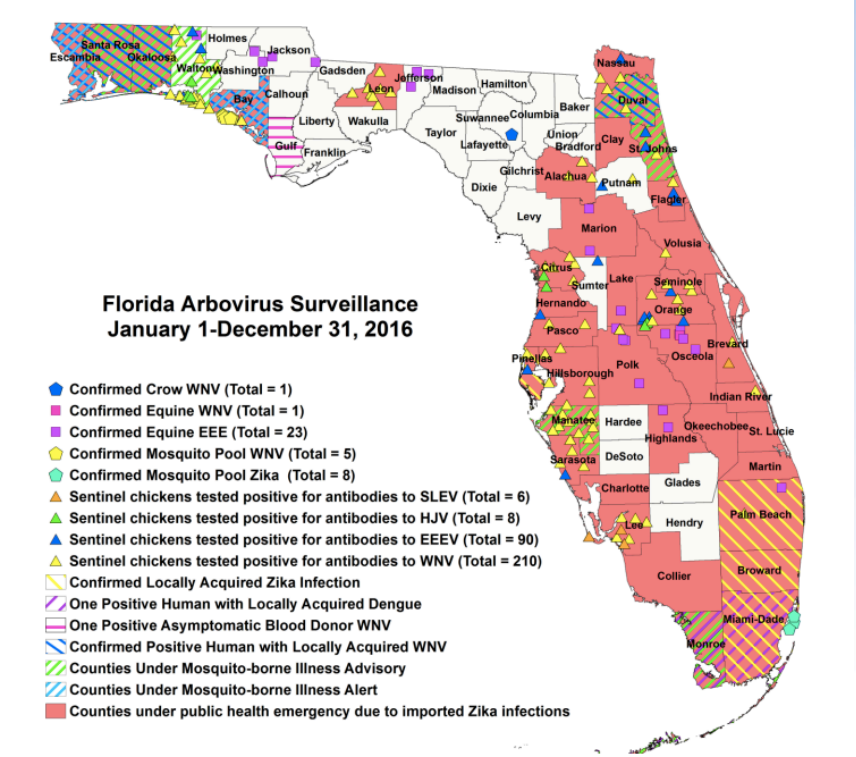
Disease Carrying (human)	Nuisance
Aedes aegypti, albopictus Zika, Chikungunya, Dengue Fever, Yellow Fever	***Aedes taeniorhynchus*** Dog heartworm
Culex nigripalpus, quinquefasciatus SLE, West Nile Virus	***Mansonia*** Dog heartworm
Anopheles spp Malaria	***Psorophora spp***
Melanoconion Everglades Virus and other potential emerging pathogens	

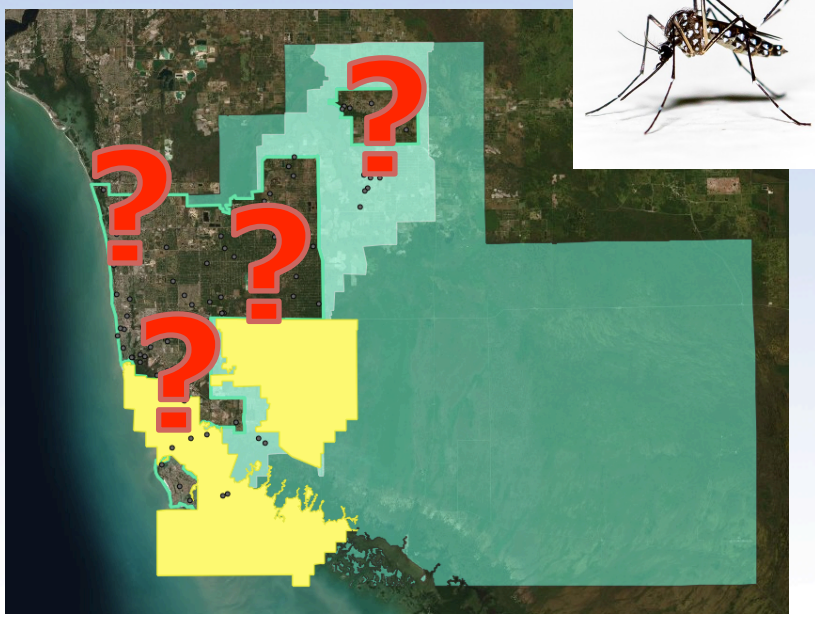
COLLIER MOSQUITO CONTROL DISTRICT

Container-inhabiting mosquitoes

- *Aedes aegypti, Aedes albopictus* and *Culex quinquefasciatus*

Container-inhabiting mosquito surveillance





COLLIER MOSQUITO CONTROL DISTRICT

Container-inhabiting mosquito surveillance

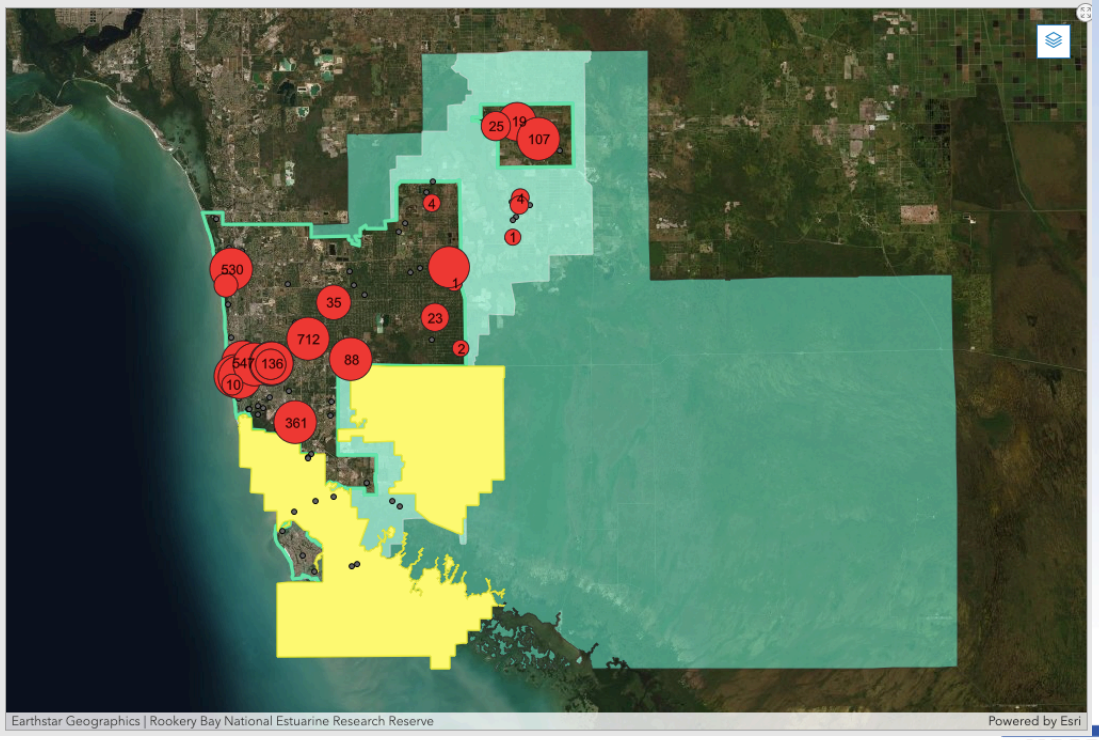
Aedes aegypti

Species counts by location | Year: 2021 | Species: Ae. aegypti

Total mosquitoes
900k
collected this year

Total of species
4.9k
collected this year

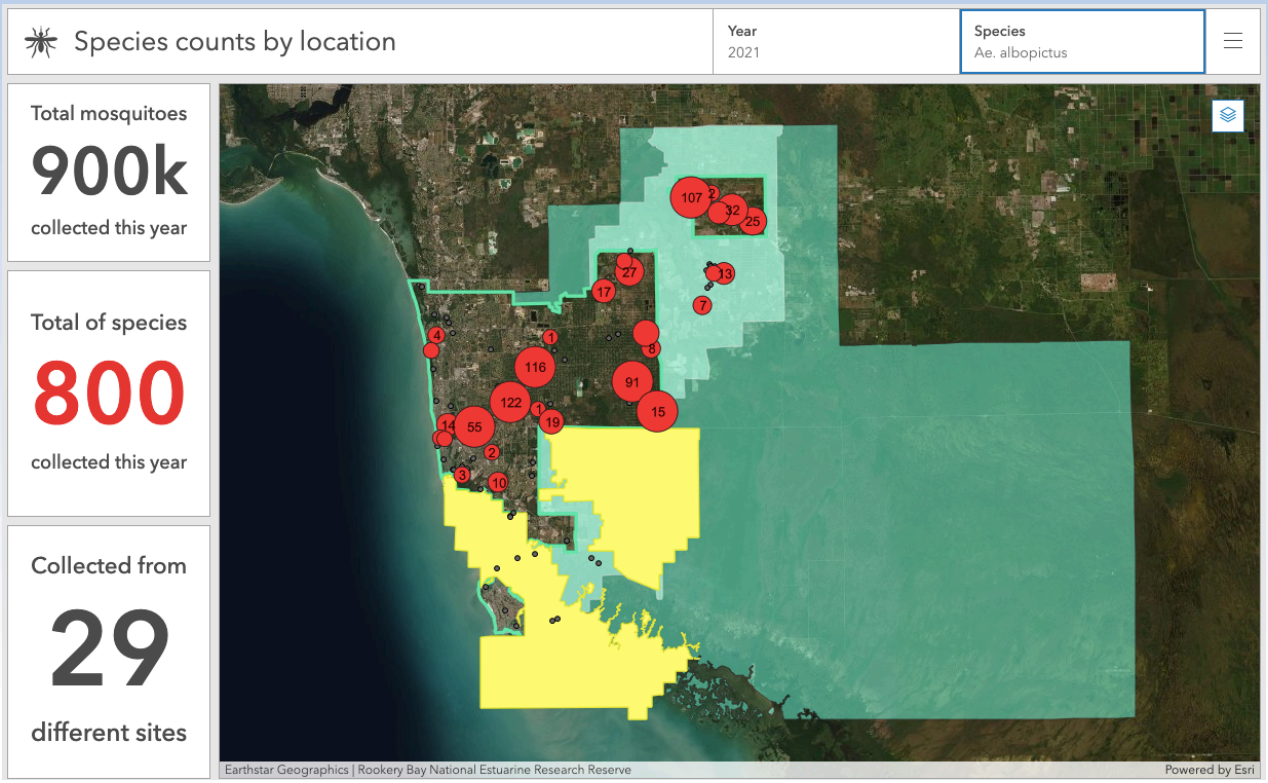
Collected from
37
different sites



COLLIER MOSQUITO CONTROL DISTRICT

Container-inhabiting mosquito surveillance

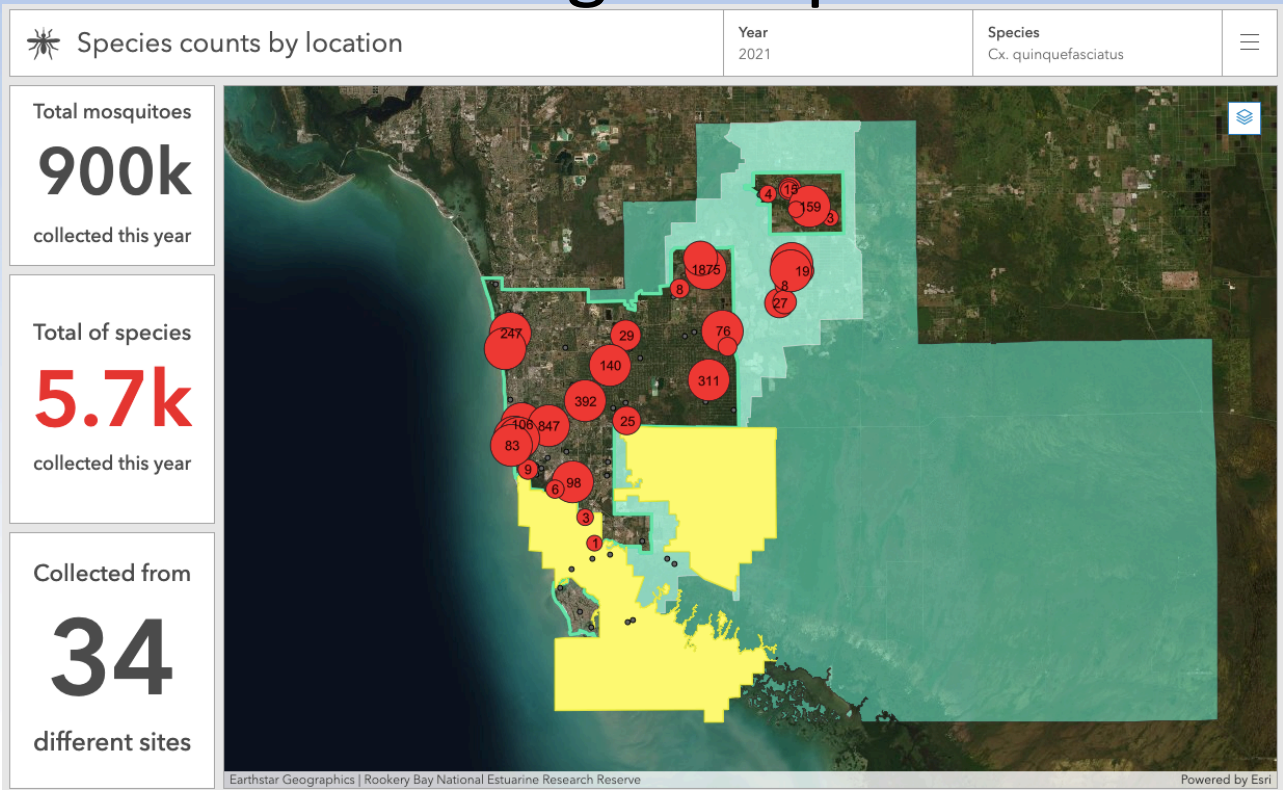
Aedes albopictus



COLLIER MOSQUITO CONTROL DISTRICT

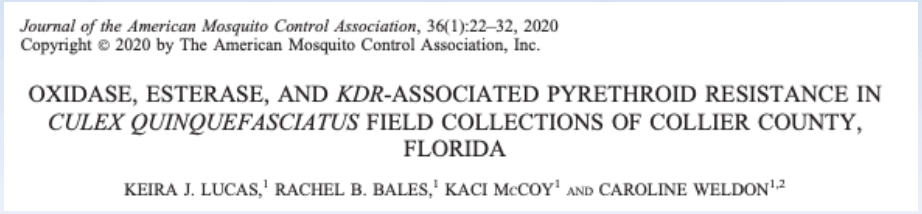
Container-inhabiting mosquito surveillance

Culex quinquefasciatus



Container-inhabiting mosquitoes in Collier

- Pyrethroid-based insecticide resistance common in Colliers ***Aedes aegypti*** (Estep *et al*. 2018; Schluep & Buckner 2021) and ***Culex quinquefasciatus*** (Lucas *et al.* 2020; Watkins *et al.* 2020)
- Naled resistance in Colliers ***Aedes aegypti*** (unpublished)



Journal of the American Mosquito Control Association, 36(1):22–32, 2020
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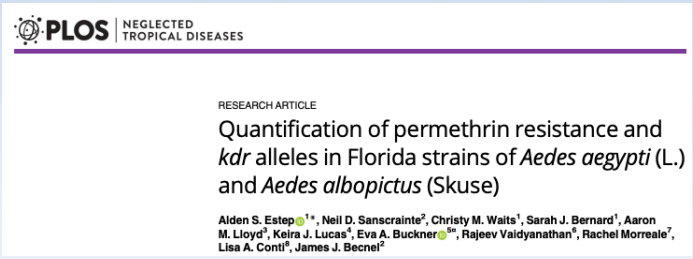
OXIDASE, ESTERASE, AND *KDR*-ASSOCIATED PYRETHROID RESISTANCE IN *CULEX QUINQUEFASCIATUS* FIELD COLLECTIONS OF COLLIER COUNTY, FLORIDA

KEIRA J. LUCAS,[1] RACHEL B. BALES,[1] KACI McCOY[1] AND CAROLINE WELDON[1,2]



PLOS | NEGLECTED TROPICAL DISEASES

RESEARCH ARTICLE

Quantification of permethrin resistance and *kdr* alleles in Florida strains of *Aedes aegypti* (L.) and *Aedes albopictus* (Skuse)

Alden S. Estep[1]*, Neil D. Sanscrainte[2], Christy M. Waits[1], Sarah J. Bernard[1], Aaron M. Lloyd[3], Keira J. Lucas[4], Eva A. Buckner[5]*, Rajeev Vaidyanathan[6], Rachel Morreale[7], Lisa A. Conti[8], James J. Becnel[2]

insects

MDPI

Article

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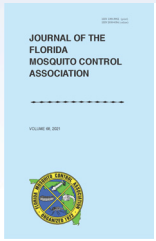
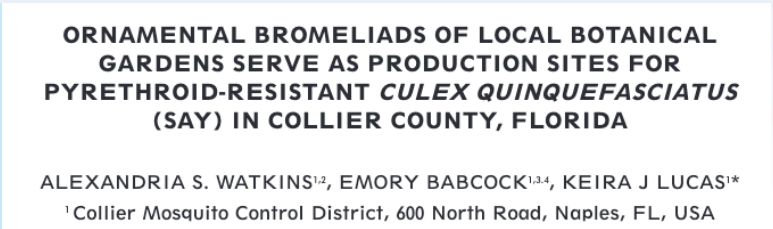
Sierra M. Schluep and Eva A. Buckner *



JOURNAL OF THE FLORIDA MOSQUITO CONTROL ASSOCIATION



ORNAMENTAL BROMELIADS OF LOCAL BOTANICAL GARDENS SERVE AS PRODUCTION SITES FOR PYRETHROID-RESISTANT *CULEX QUINQUEFASCIATUS* (SAY) IN COLLIER COUNTY, FLORIDA

ALEXANDRIA S. WATKINS[1,2], EMORY BABCOCK[1,3,4], KEIRA J LUCAS[1]*

[1] Collier Mosquito Control District, 600 North Road, Naples, FL, USA

COLLIER MOSQUITO CONTROL DISTRICT

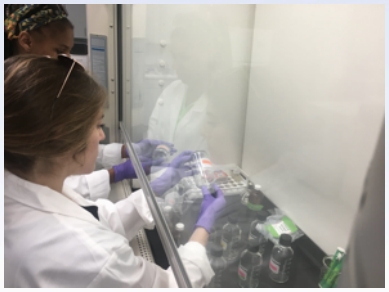
Laboratory Evaluation - CDC Bottle Bioassay

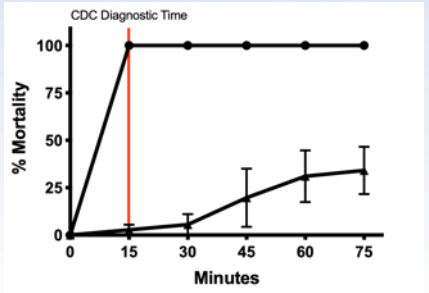
Coat bottles with known amount (diagnostic dose) of control material.

Allow material to dry. Add 20-25 adult mosquitoes per bottle (3-4 replicates).

Collect data and calculate percent mortality every 15 minutes for 2 hours.

Create mortality curves and determine susceptibility status at CDC diagnostic time.





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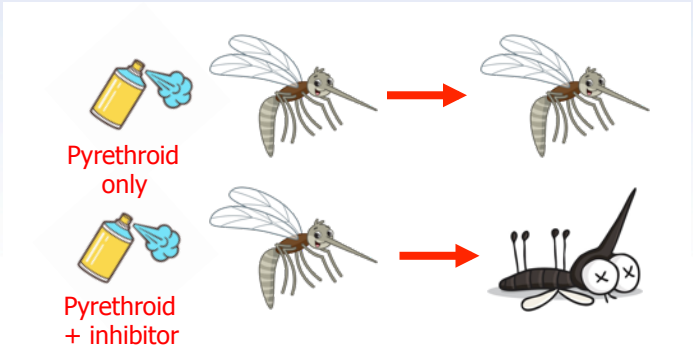
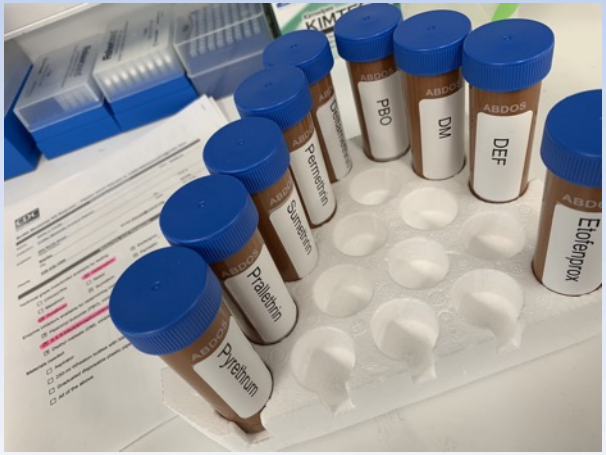
What factors contribute to resistance?

- *kdr* genotyping
- *kdr* phenotyping
- Inhibitors
  - PBO: oxidase
  - DEF: esterase
  - DEM: glutathione transferases





CDC
CENTERS FOR DISEASE CONTROL AND PREVENTION

COLLIER MOSQUITO CONTROL DISTRICT

Laboratory Evaluation – *kdr* phenotyping/Recovery Assays

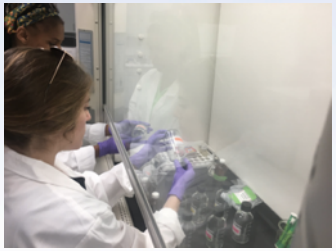
Coat bottles with known amount (diagnostic dose) of control material.

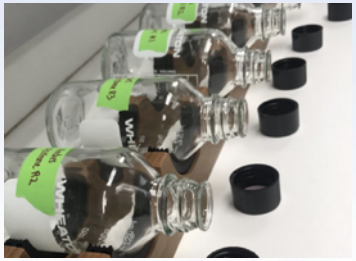
Allow material to dry. Add 20-25 adult mosquitoes per bottle (3-4 replicates).

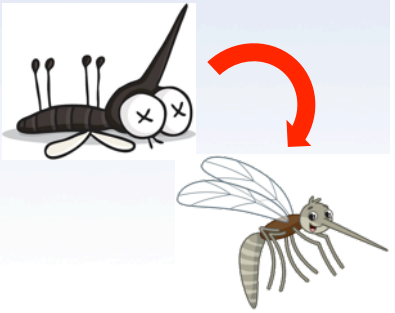
Expose mosquitoes to insecticide for 2 hrs. Calculate mortality after 2 hrs.

Transfer "dead" and alive mosquitoes to holding cages. Calculate mortality after 24 hrs.

Determine recovery rates. Recovery = resistance due to mutation.







COLLIER MOSQUITO CONTROL DISTRICT

Laboratory Evaluation – Metabolic Assays

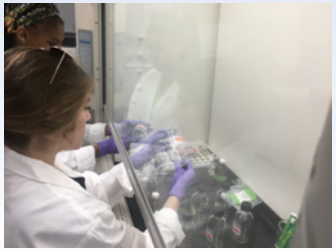
Coat bottles with known amount of inhibitor. Dry for 1 hr.

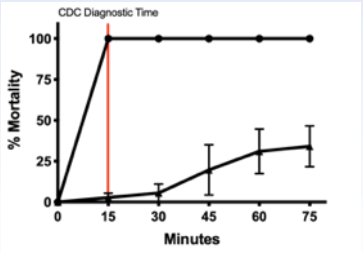
Expose mosquitoes to inhibitor for 1 hr.

Rest mosquitoes for 1 hr.

Perform CDC bottle bioassay as normal.

Determine susceptibility status at CDC diagnostic time.





Does this resistance translate to resistance in the field?
Ground Field Cage Trials

Select initial application rate for testing and calculate dilution requirements and flow rates.

Set up sampling stations: cages with 20-25 adult mosquitoes and slide impingers (4 replicates – 15 ft apart).

Treat mosquitoes using hand fogger – 10 to 20 feet from sampling station.

Bring back to lab. Collect data and calculate percent mortality every 15 mins for 2 hours, 8 hrs and 24 hrs post treatment.

Create mortality curves and determine treatment efficacy.

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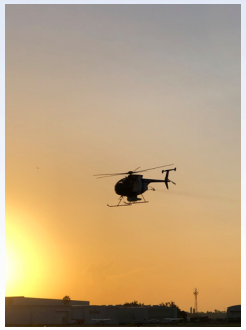
Does this resistance translate to resistance in the field?
Aerial Field Cage Trials

Select initial application rate for testing, calibrate and droplet check.

Set up sampling stations with cages with 20-25 adult mosquitos within treatment area in 3x3 grid (9 total, 100 feet apart).

Complete "treatment block" over caged mosquitoes.



Bring back to lab. Collect data and calculate percent mortality every 15 mins for 2 hours, 8 hrs and 24 hrs post treatment.

Create mortality curves and determine treatment efficacy.

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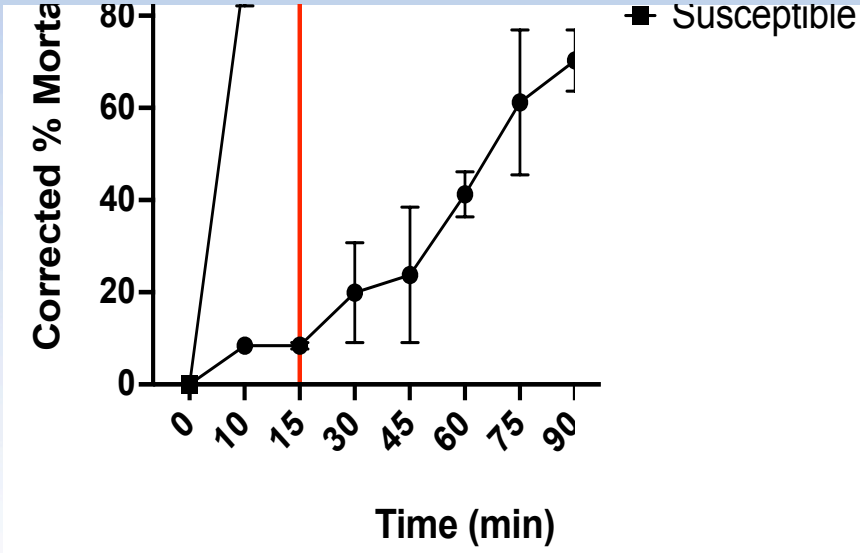
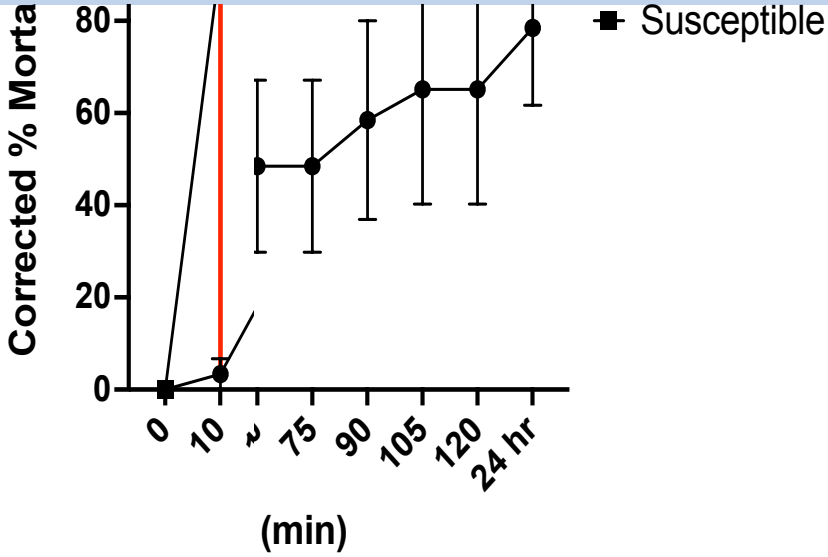
Insecticide Susceptibility of *Aedes aegypti*



RESISTANT



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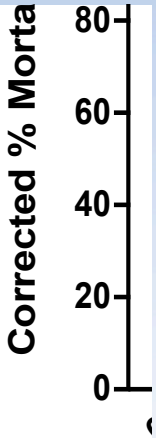

RESISTANT

CDC Bottle Bioassays – October 2021 (Collier Strain)

COLLIER MOSQUITO CONTROL DISTRICT

Pyrethroid Susceptibility of *Aedes aegypti*

PLOS | NEGLECTED TROPICAL DISEASES

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kdr mutations F1534C and V1016I
Esterase activity

Bottle bioassay, metabolic assays, *kdr* genotyping, and field cage trials

insects MDPI

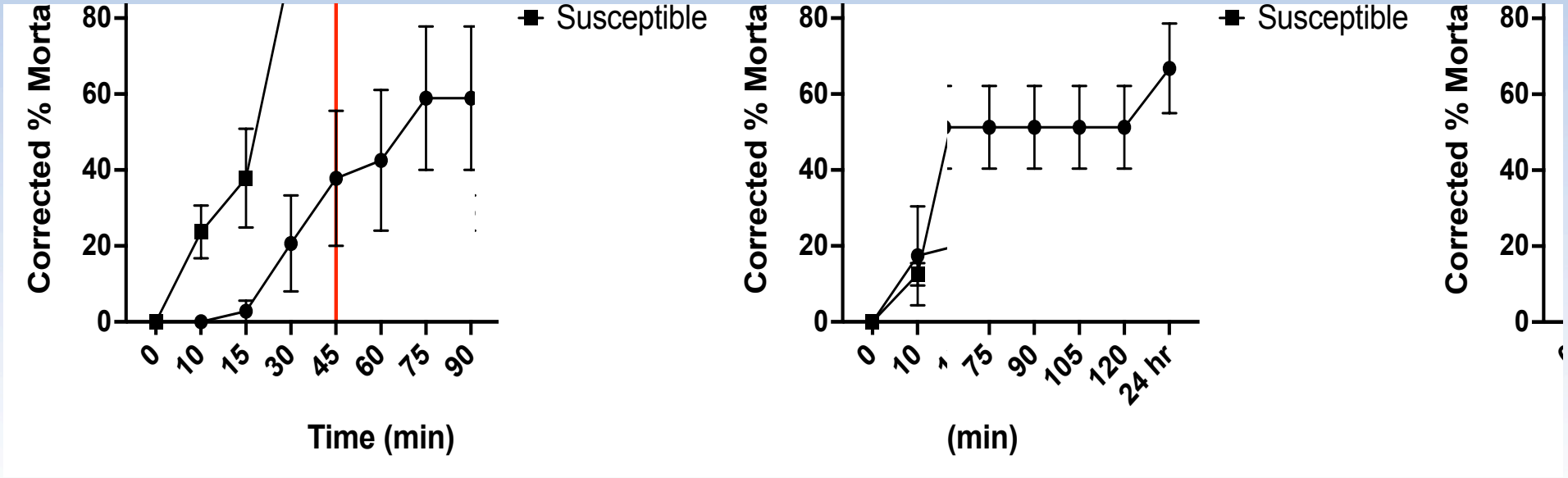
Article

Metabolic Resistance in Permethrin-Resistant Florida *Aedes aegypti* (Diptera: Culicidae)

Sierra M. Schluep and Eva A. Buckner *

COLLIER MOSQUITO CONTROL DISTRICT

Insecticide Susceptibility of *Culex quinquefasciatus*



RESISTANT **RESISTANT** **SUSCEPTIBLE**

CDC Bottle Bioassays – October 2021 (Big Cypress Elementary)

COLLIER MOSQUITO CONTROL DISTRICT

Pyrethroid Susceptibility of *Culex quinquefasciatus*

Journal of the American Mosquito Control Association, 36(1):22–32, 2020

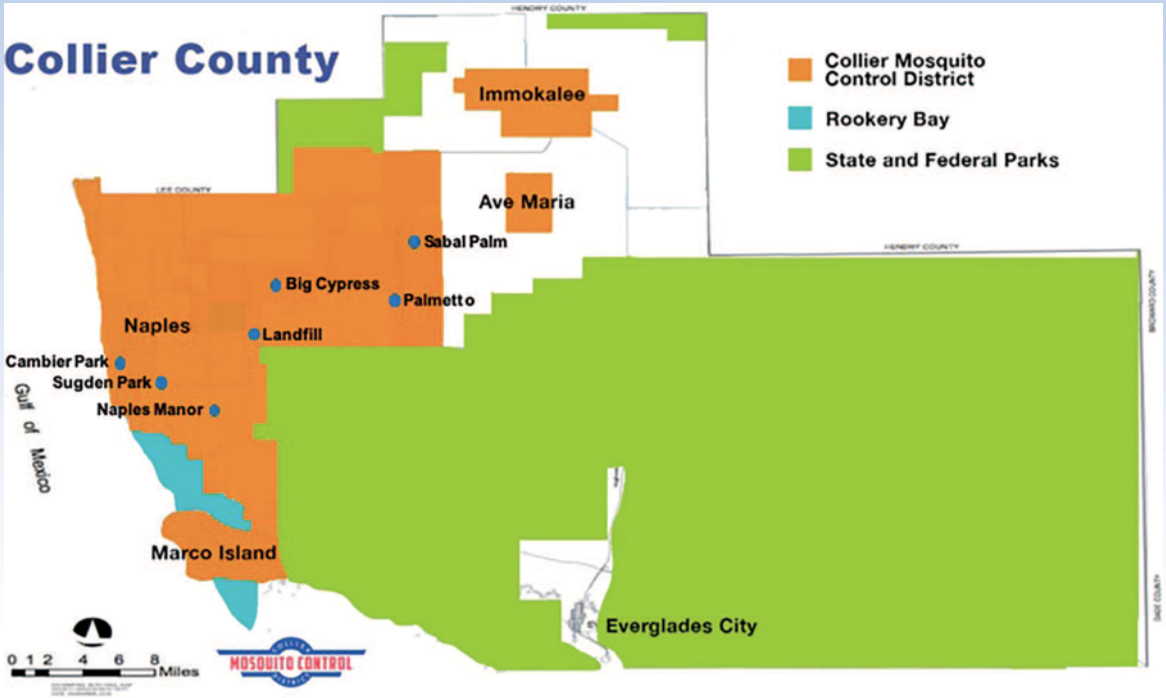
OXIDASE, ESTERASE, AND *KDR*-ASSOCIATED PYRETHROID RESISTANCE IN *CULEX QUINQUEFASCIATUS* FIELD COLLECTIONS OF COLLIER COUNTY, FLORIDA

KEIRA J. LUCAS,[1] RACHEL B. BALES,[1] KACI McCOY[1] AND CAROLINE WELDON[1,2]

Bottle bioassay, metabolic assays, recovery assays, *kdr* genotyping, and field cage trials

kdr mutations L1014F
Oxidase activity
Esterase activity

Field Collections

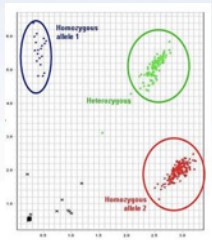


Lucas et al 2020 (JAMCA)

Cx. quinquefasciatus kdr Mutation

DNA Extraction

PCR



Analysis

Location	Habitat	Genotypic Frequency			Allele Frequency	
		LL	*LF*	*FF*	*L*	*F*
Cambier Park	Storm Drain	.2051	.5385	.2564	.4744	.5256
Sugden Park	Containers Storm Drain	.2564	.5641	.1795	.5385	.4615
Naples Manor	Containers	.3409	.5455	.1136	.6136	.3864
Landfill	Containers Storm Drain	.4500	.5000	.0500	.7000	.3000
Big Cypress Elementary	Septic	.5313	.4062	.0625	.7344	.2656
Palmetto Elementary	Septic	.7692	.2308	0	.8846	.1154
Sabal Palm Elementary	Septic	.8788	.1212	0	.9394	.0606

Decreasing mutation frequency

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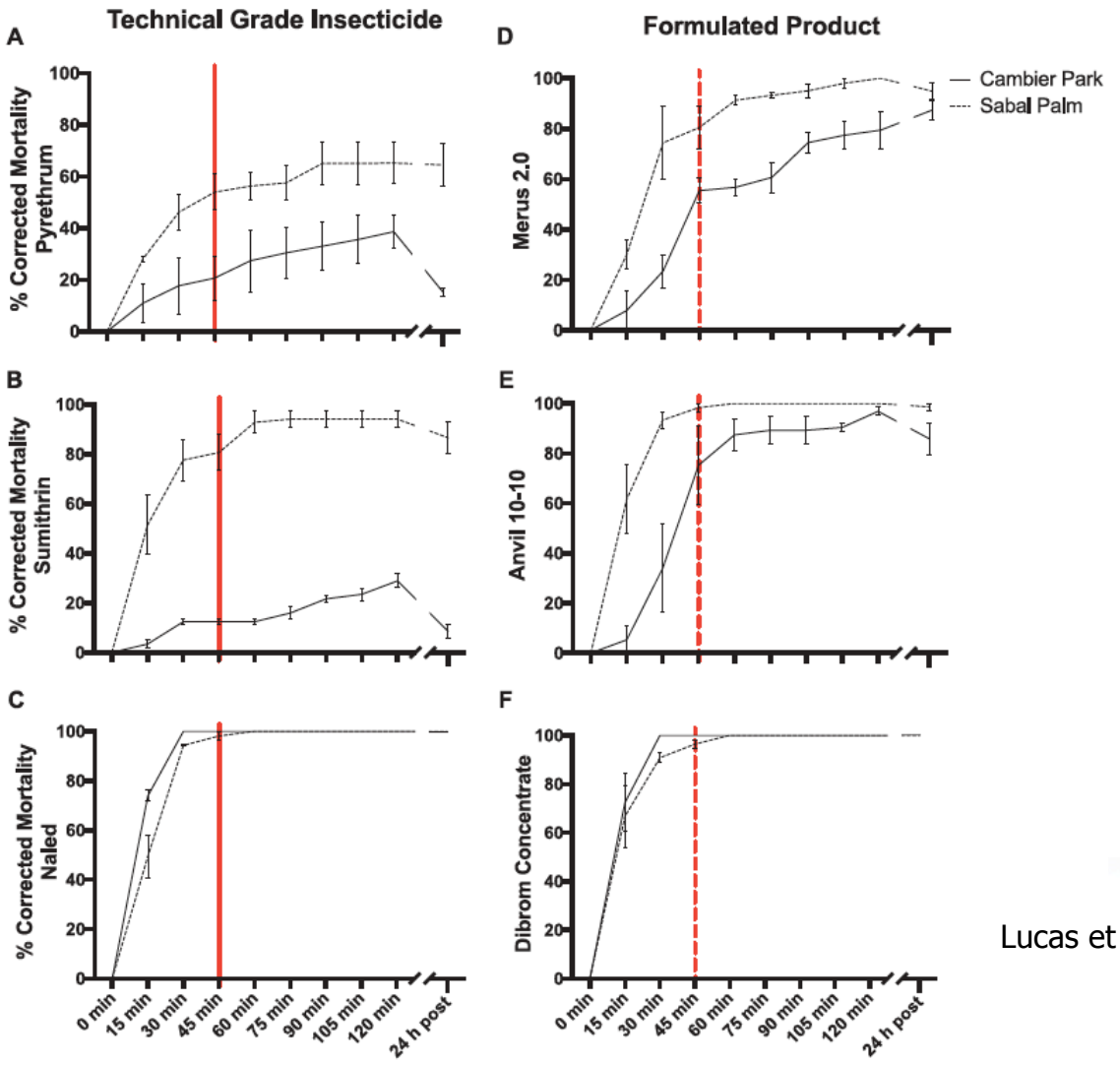

Cambier Park – High level of pyrethroid resistance

Sabal Palm – Lower level of pyrethroid resistance

No resistance to Naled/ Dibrom

Lucas et al 2020 (JAMCA)

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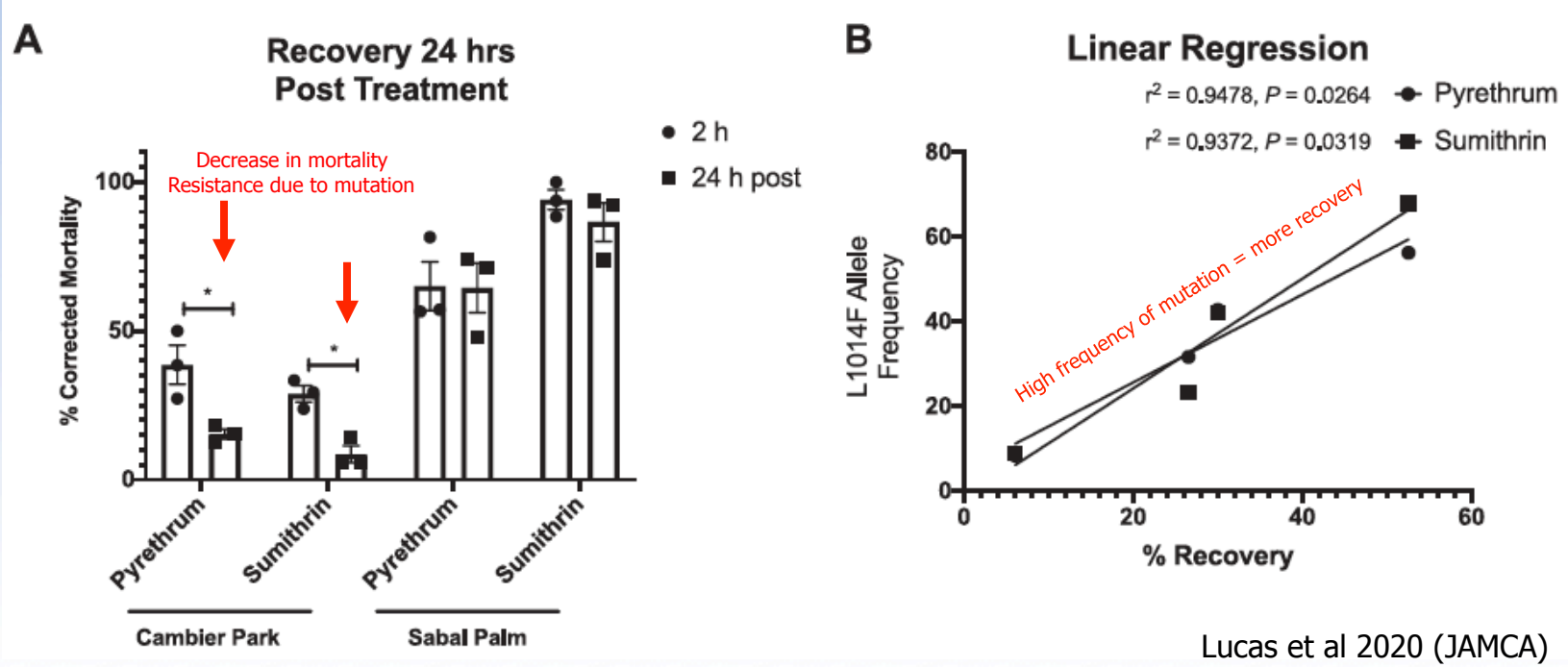
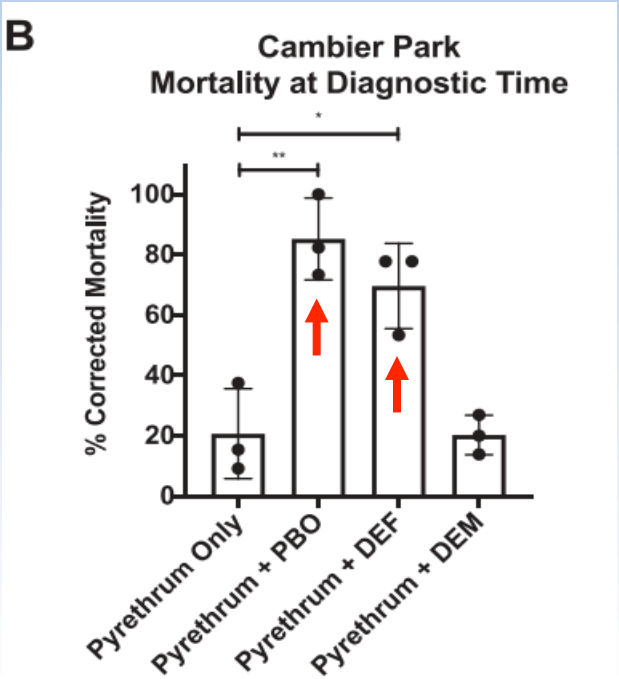
How much of this resistance is attributed to the mutation?

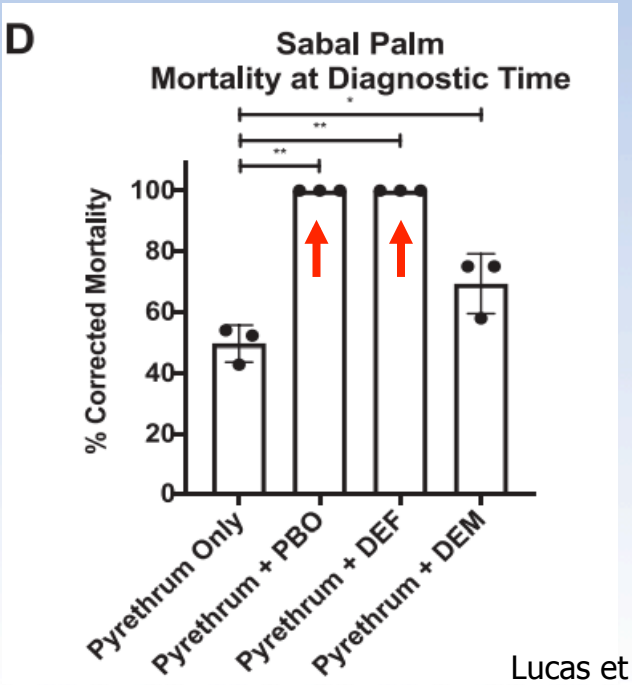


Lucas et al 2020 (JAMCA)

The mutation plays a role, at least in part, to the pyrethroid resistance status of our *Culex* mosquitoes.

What other factors are resulting in resistance?

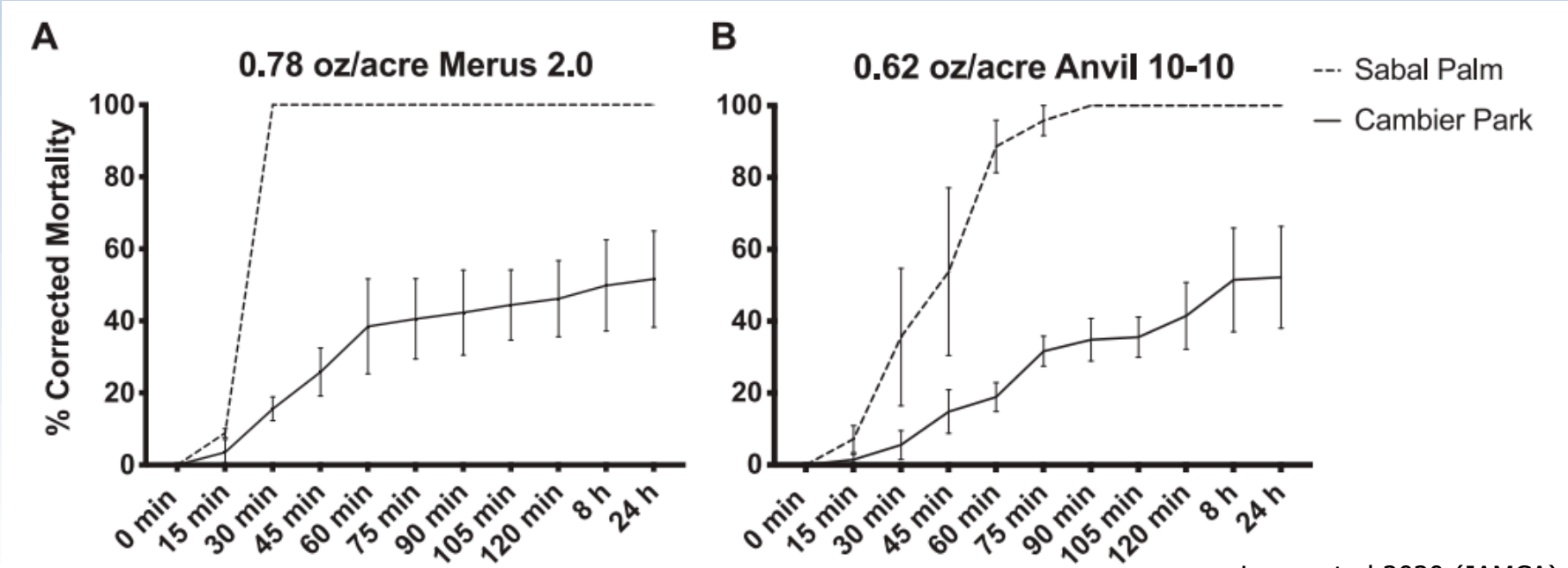




Lucas et al 2020 (JAMCA)

Oxidase and esterase metabolism contribute the majority of resistance in our *Culex* mosquitoes

Does this resistance translate to resistance in the field?
Ground Field Cage Trials



Lucas et al 2020 (JAMCA)

Cambier Park = High frequency of mutation = Reduced efficacy of product
Sabal Palm = Low frequency of mutation = 100% knockdown

COLLIER MOSQUITO CONTROL DISTRICT

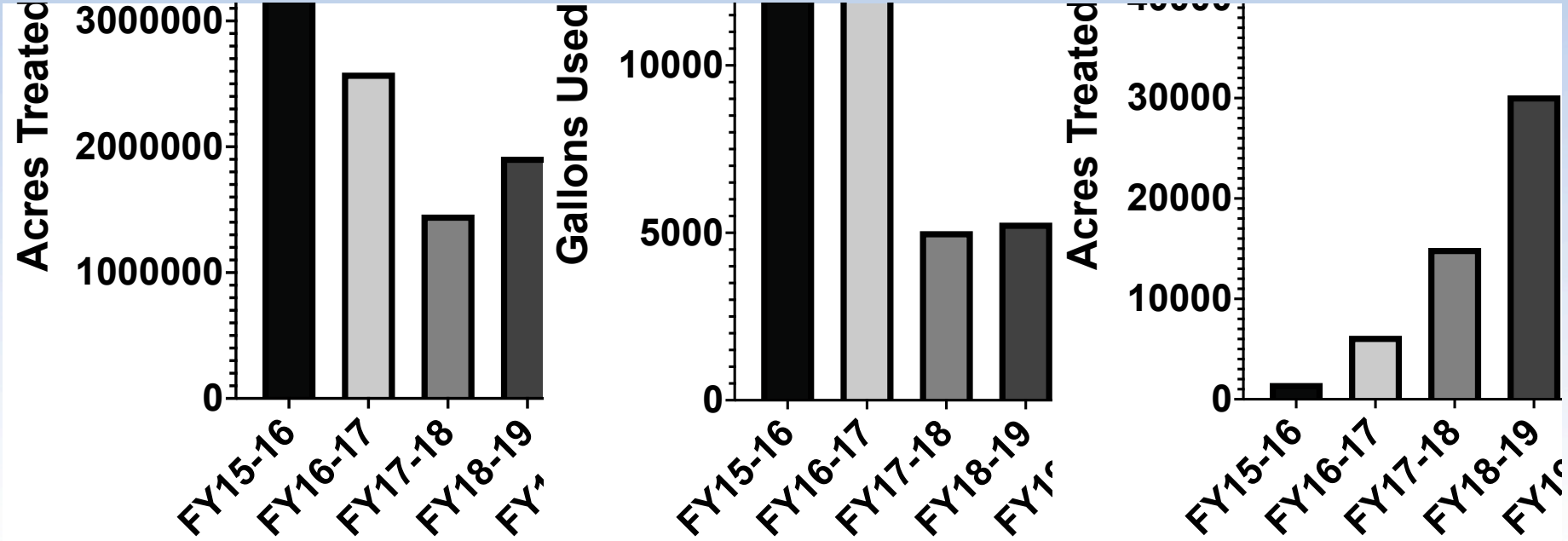
COLLIER MOSQUITO CONTROL DISTRICT

Conventional Flat Fan

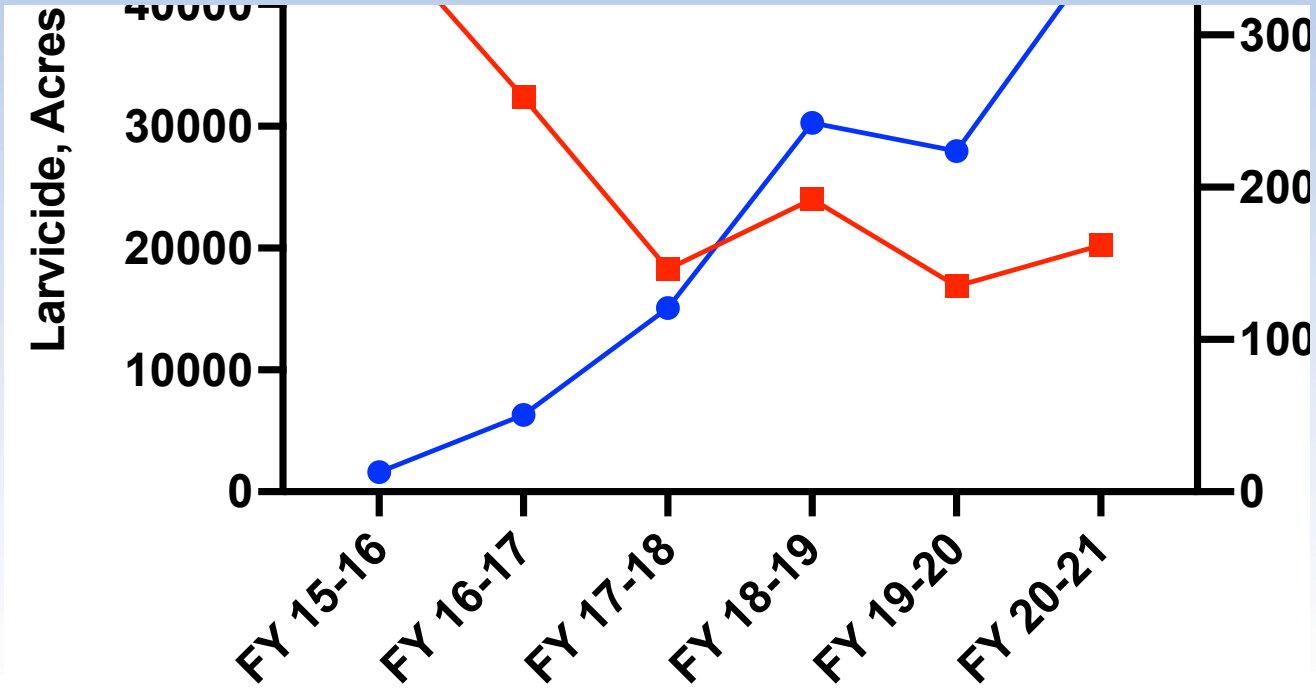
High Pressure Hydraulic

Rotary Atomizers

Mosquito Control Then vs. Now



Mosquito Control Then vs. Now



Adulticide at CMCD

DIBROM CONCENTRATE

clarke DUET® HD Dual-Action Adulticide

clarke MERUS® 3.0

WISDOM TC FLOWABLE

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Liquid larvicide applications at CMCD

- Buffalo Turbine & Liquid Larvicide unit for rotary wing aircraft in 2016
- A1 Superduty and two A1 Rangers in 2019 to expand capabilities
- UAV applications in 2019 for fine-tuned applications of smaller treatment areas
- A1 Superduty in 2021

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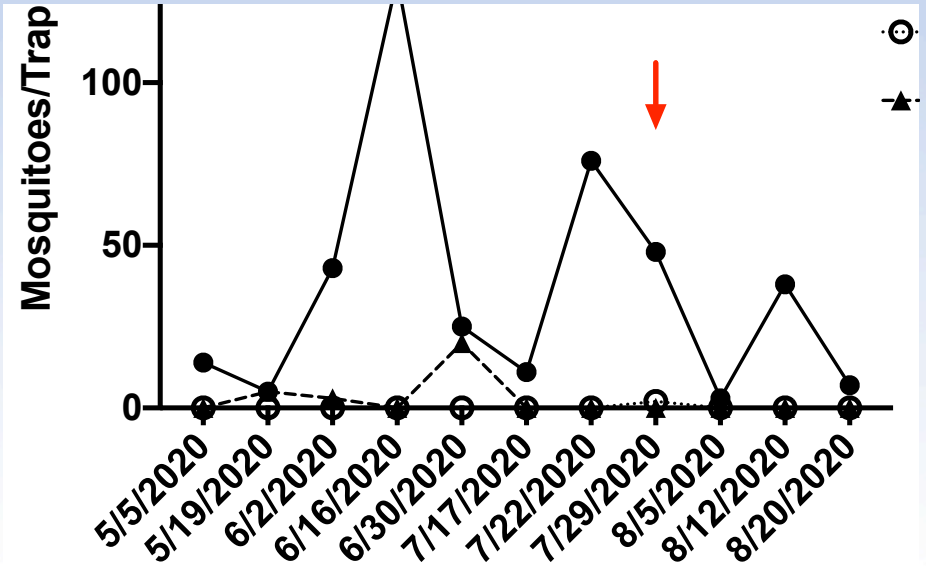
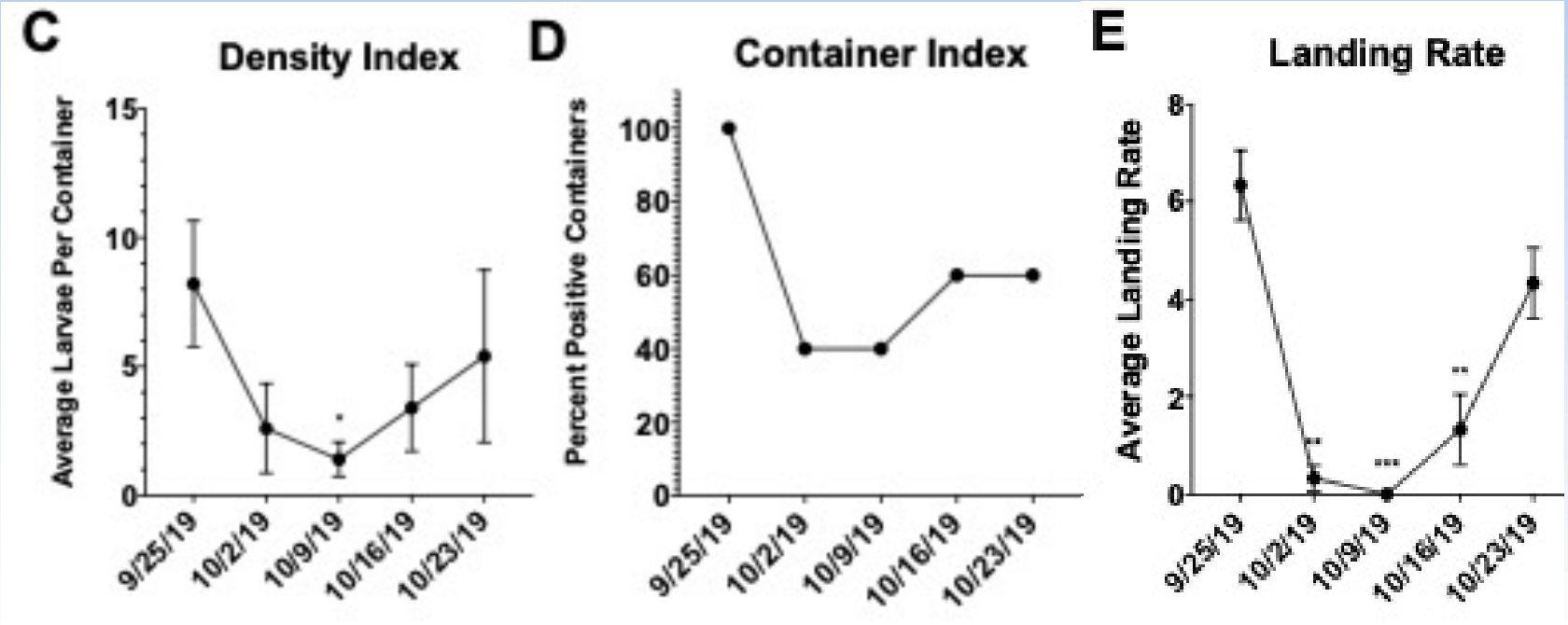
Truck-based WALS applications

- **Vectobac WDG (Water Dispersible Granule)** applied at 0.5 lbs/acre

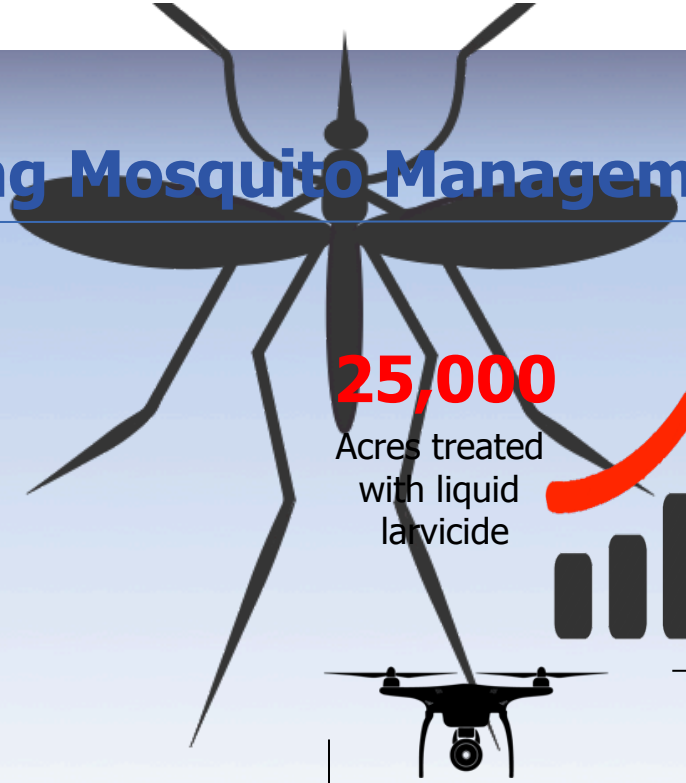


Drone-based WALS applications



Lucas & Brake et al 2020. (JFMCA)

Container Inhabiting Mosquito Management



62%

Gallons of organophosphates used since 2016

3,352

Students reached in classrooms during 2019

25,000

Acres treated with liquid larvicide

Pyrethroid & naled resistance identified in container species

1400

Mosquito pools tested for VBD

9k+

Mosquitofish distributed in 2021

OMRI

OMRI-listed control materials

UAS

WALS Applications

1500

Trap collections

COLLIER MOSQUITO CONTROL DISTRICT

Thank you

Collier Mosquito Control District

- Executive Director: **Patrick Linn, MS, MSHAPI**
- Director of Research: **Rebecca Heinig, PhD**
- Director of Operations: **Nate Phillips**
- Director of Technical Development: **Peter Brake**
- Director of Communications: **Robin King**
- Field Technician Supervisor: **Richie Ryan**
- Biologist: **Rachel Bales**
- Logistics Coordinator: **Sara Grant**

Valent Biosciences

- Leanne Lake
- Candace Royals
- Banu Kesavaraju
- Carolina Torres Gutierrez

Questions

Keira J. Lucas, PhD
Deputy Executive Director
E-mail: klucas@cmcd.org

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