EVALUATION OF CDC LIGHT TRAP, BG SENTINEL TRAP, AND MMX TRAP FOR THE COLLECTION OF SALT MARSH MOSQUITOES IN ANASTASIA STATE PARK, SAINT AUGUSTINE, FLORIDA

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ABSTRACT

Salt marsh mosquitoes are major nuisance pests during the periods of high mosquito activity, especially after major storm events. In 2016-2017, Saint John's County, Florida, USA was struck by two major hurricanes that resulted in multiple outbreaks of salt marsh mosquito populations. To optimize the surveillance of two salt marsh mosquitoes, (*Aedes taeniorhynchus* and *Ae. sollicitans*, three types of traps (the Centers for Disease Control (CDC) Light trap, Bio-gents Sentinel (BG) trap and Counter Flow Geometry Model (MMX) trap were tested for their capacity to capture the highest numbers of high quality live specimens for laboratory bioassays. Each trap type was tested in Anastasia State Park, located along a major salt marsh area in Saint John's County. Although the MMX trap captured most of the salt marsh mosquitoes collected, the numbers of mosquitoes captured was not statistically significant compared to the other trap types. However, there was a significant difference in the numbers between *Ae. taeniorhynchus* and *Ae. sollicitans* in the MMX traps. The MMX trap is preferred for capturing salt marsh mosquitoes that are in high quality for the CDC bottle bioassays.

Key Words: *Aedes taeniorhynchus*, *Aedes sollicitans*, surveillance, salt marsh, CDC light trap

Salt marsh mosquitoes are nuisance pests to Florida residents and tourists due to their aggressive biting behavior and long flight range (Rey et al. 2012; Nayar 1985; Hribar et al. 2010). These mosquitoes are also of a public health concern to people due to allergic reactions caused by their bites and their potential as vectors of disease pathogens such as the dog heartworm and Venezuelan Eastern Encephalitis virus, both to dogs and residents in Saint John's County, Florida, USA, respectively (Peng et al. 2004; Sudia et al. 1971; Weaver et al. 1996). It was noted that seasonality, temperature, flood conditions and hurricanes are the major factors that affecting egg hatching rates, which result in noticeable increases in populations of salt marsh mosquitoes (Hribar et al. 2010).

Two salt marsh mosquito species, *Aedes taeniorhynchus* (Weidemann) and *Ae. sollicitans* (Walker) are primarily targeted by Anastasia Mosquito Control District (AMCD),

St. Augustine, FL to control. *Aedes taeniorhynchus*, also known as the Black Salt Marsh mosquito, breeds in brackish water in North, Central, and South America; while *Ae. sollicitans*, also known as the Eastern Salt Marsh mosquito, is found in saline wetlands, dense salt marshes, shallow pools, and inland in collected brackish water. In the 2016-2017, Saint John's County was hit by thunderstorms throughout the summer and two hurricanes (Matthew and Irma) that accounted for over 56 cm of rain levels. Over 200 service requests per day were raised from the residents living near salt marsh habitats in Anastasia Island. Accordingly, and in prompt response, AMCD sent multiple teams of technicians to treat the island using insecticides, like Mosquito Mist and Aqualuer 20-20 at night and Talstar-P and DUET during the day. However, the repeated and intensive treatments against the salt marsh mosquito populations raised concerns of the potential risk of insecticide resistance development in

the targeted mosquito populations. To test for insecticide resistance in the salt marsh mosquito populations, AMCD used the CDC bottle bioassay, a method that determines the time required for an insecticide to affect

a mosquito from initial exposure to knock-down and then final death.

Optimized trapping protocols are paramount for the CDC bottle bioassay due to the large numbers of intact live mosquitoes

Figure 1. Placement of each MMX trap, CDC light trap, and BG trap. Each replicate (Rep) is encircled by a grey polygon. Within each Rep, the blue blips represent the positions of MMX traps, the purple blips represent the positions of CDC light traps, and the orange blips represent the positions of BG traps. The black scale bar on the right bottom represents 17 meters.

needed to test for susceptibility/resistance bioassays. Most of the previous research has focused on the capture of salt marsh mosquitoes for studying attractant combinations for the CDC light trap (Kline and Lemire, 1995; Rueda et al. 2001). However few studies had been conducted to assess the differences in capture rate of salt marsh mosquitoes by multiple trap types (Smith et al. 2016). Three types of traps, the Centers for Disease Control (CDC) Light trap, Biogents Sentinel (BG) trap and the Counter Flow Geometry Model (MMX) trap were used in this study for salt marsh mosquito collections. This was with the purpose to test the differential capacity of these traps for capturing high quality live salt marsh mosquitoes for lab bioassay.

Three of each trap type were set at a picnic area in Anastasia State Park (29.866186 N, 81.272030 W), and each trap was placed at 9.1–36.6 m apart. All traps were baited with dry ice (carbon dioxide source) as an attractant, but the CDC traps (John W. Hock Company, Gainesville, FL, USA) and BG traps (Biogents AG, Regensburg, Germany) were also baited with Octenol (Biosensory, Putnam, CT, USA) and BG lure (ADAPCO, Sanford, FL, USA), respectively. The MMX traps (American Biophysics Corp., RI) and CDC traps were hung with shepherd’s hooks at one m above ground, while the BG traps were placed on the ground. CDC traps were hooked up to 6-volt batteries; while the BG and MMX traps were hooked up to 12-volt batteries.

All traps were set in the afternoon and collected the next day. All specimens from each trap were identified to species and counted to determine the trap that collected the highest abundance of salt marsh mosquitoes. A goodness-of-fit test was used to determine that the datasets conformed to

non-normal, heteroscedastic behavior. Thus, a non-parametric Kruskal-Wallis test was applied to the data to determine if trap capture differences were statistically significant.

The abundance of the two salt marsh mosquitoes, *Ae. taeniorhynchus* and *Ae. sollicitans* captured in the MMX trap, CDC Light trap, and BG trap was analyzed and presented in Table (1). A total of 697 *Ae. taeniorhynchus* and 154 *Ae. sollicitans*, with an average across all three replicates at 232 *Ae. taeniorhynchus* and 51 *Ae. sollicitans* were captured in the MMX trap. In the CDC light trap, a total of 499 *Ae. taeniorhynchus* and 149 *Ae. sollicitans*, with an average across all three replicates at 166 *Ae. taeniorhynchus* and 50 *Ae. sollicitans* were captured. The BG trap captured the lowest number of mosquitoes, with a total of 16 *Ae. taeniorhynchus* and 39 *Ae. sollicitans* and an average across all three replicates at five *Ae. taeniorhynchus* and 13 *Ae. sollicitans*. Although it seemed to perform well, the mosquito numbers captured by the MMX traps were not statistically significant compared to those captured by the CDC light traps and BG traps. However, there was a significant difference in abundance between *Ae. taeniorhynchus* and *Ae. sollicitans* in the MMX trap, though the difference is marginal ($N = 6$, $\chi^2 = 3.8571$, $DF = 4$, $P = 0.0497$).

Despite the comparable salt marsh mosquitoes capturing efficacy between the three tested trap types, the MMX trap is preferred for capturing live mosquitoes suitable for the CDC bottle bioassay, due to its counter flow updraft system, which collects mosquitoes unharmed in the clear plastic capture chamber. Unlike the MMX traps, CDC light traps suck mosquitoes into capture jars using suction force generated through miniature fans. The mosquitoes contact the suction fan blades upon capture, which potentially dam-

Table 1. Average numbers of *Aedes taeniorhynchus* and *Ae. sollicitans* captured in the MMX trap, CDC light trap, and BG trap.

Species	MMX trap Average	CDC light traps Average	BG traps Average
<i>Aedes taeniorhynchus</i>	232.3 ± 83.5 ^{Ba}	166.3 ± 126.9 ^{Aa}	5.3 ± 3.5 ^{Aa}
<i>Ae. sollicitans</i>	51.3 ± 19.5 ^{Aa}	49.7 ± 27.1 ^{Aa}	13.0 ± 9.1 ^{Aa}

N = 3 traps.
Lowercase superscript letters indicate no significant difference for total species between trap types.
Uppercase superscript letters indicate a significant difference in abundance between the two species (P-value = 0.0497).

ages major appendages and thus confounds bottle bioassays. The CDC bottle bioassay detects mosquito susceptibility levels or resistance to active ingredients in insecticides by measuring their morbidity and mortality over designated exposure time (CDC, 2010). Damage to mosquitoes before testing can have a significant impact on the rate of mortality in control and treatment groups. For this reason, minimizing damage to mosquitoes during trap capture is a major consideration when collecting them from the field.

Although the mortality of mosquitoes caught in CDC light traps and MMX traps was not analyzed, future experiments could look at how the two suction systems affect downstream applications from field collected specimens. Future experiments could also test alternative attractants and new trap types for improved capture rates of *Ae. taeniorhynchus* and *Ae. sollicitans*. This study, ultimately, analyzed the capacity of different traps for capturing a high numbers of live undamaged salt marsh mosquitoes not only for insecticide susceptibility /resistance bioassays, but also for lab colony development, molecular testing, and other experiments that increase our understanding biological and ecology of the collected mosquito species for informed and targeted control operations.

ACKNOWLEDGEMENTS

We would like to thank Mr. Jim Cilek and James Richardson from the USDA/ CMAVE for their help with statistical analyses for this study.

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