Journal of the American Mosquito Control Association, 31(1):26–31, 2015

COMPARISON OF BG-SENTINEL® TRAP AND OVIPOSITION CUPS FOR *AEDES AEGYPTI* AND *AEDES ALBOPICTUS* SURVEILLANCE IN JACKSONVILLE, FLORIDA, USA

JENNIFER A. WRIGHT,[1] RYAN T. LARSON,[1] ALEC G. RICHARDSON,[1] NOEL M. COTE,[1] CRAIG A. STOOPS,[2] MARAH CLARK[3] AND PETER J. OBENAUER[1]

ABSTRACT. The BG-Sentinel® (BGS) trap and oviposition cups (OCs) have both proven effective in the surveillance of *Aedes* species. This study aimed to determine which of the 2 traps could best characterize the relative population sizes of *Aedes albopictus* and *Aedes aegypti* in an urban section of Jacksonville, FL. Until 1986, *Ae. aegypti* was considered the dominant container-breeding species in urban northeastern Florida. Since the introduction of *Ae. albopictus*, *Ae. aegypti* has become almost completely extirpated. In 2011, a resurgence of *Ae. aegypti* was detected in the urban areas of Jacksonville; thus this study initially set out to determine the extent of *Ae. aegypti* reintroduction to the area. We determined that the BGS captured a greater number of adult *Ae. aegypti* than *Ae. albopictus*, while OCs did not monitor significantly different numbers of either species, even in areas where the BGS traps suggested a predominance of one species over the other. Both traps were effective at detecting *Aedes* spp.; however, the BGS proved more diverse by detecting over 20 other species as well. Our results show that in order to accurately determine vectorborne disease threats and the impact of control operations on these 2 species, multiple trapping techniques should be utilized when studying *Ae. aegypti* and *Ae. albopictus* population dynamics.

KEY WORDS *Aedes aegypti*, *Aedes albopictus*, BG-Sentinel, oviposition cup surveillance, Florida, dengue

INTRODUCTION

Dengue fever remains a serious mosquitoborne disease in many tropical and subtropical areas around the world (WHO 2009, Bhatt et al. 2013). In the USA, the principal mosquito vector, *Aedes aegypti* (L.), has been established throughout the Southeast for over 200 years (Damal et al. 2013). A second mosquito vector, *Ae. albopictus* (Skuse), was first recorded in the state of Florida (FL) in 1986 at a waste tire site in Jacksonville (O'Meara et al. 1992). While the role of *Ae. aegypti* in the transmission of dengue virus is well known, the importance of *Ae. albopictus* in the transmission of arboviruses in the USA, including dengue, is not clear. Specifically, Cache Valley virus, eastern equine encephalitis virus, Jamestown Canyon virus, La Crosse virus, and West Nile virus have been isolated from *Ae. albopictus* populations in the USA (Moore and Mitchell 1997, Gerhardt et al. 2001, Turell et al. 2005). Additionally, dengue virus has been isolated in populations of *Ae. albopictus* in the Americas (Ibanez-Bernal et al. 1997).

Both species use natural and artificial containers for larval development. Until 1986, *Ae. aegypti* was considered the dominant urban species in the southeastern United States and the subject of unsuccessful eradication efforts due to its role in yellow fever virus transmission (Schliessmann and Magennis 1964, O'Meara et al. 1993). The introduction of *Ae. albopictus* into the USA has led to *Ae. aegypti* becoming almost completely extirpated across its former range (Sibal 1994). Additionally, we confirmed this trend for the city of Jacksonville with the local mosquito control division, the Jacksonville Mosquito Control District (JMCD, Marah Clark, personal communication). Several hypotheses, including larval competition and satyrization, may explain the displacement of *Ae. aegypti* (Bargielowski and Lounibos 2014); however, in areas such as Manatee County, FL, populations of both species have been found to exist in equilibrium (Braks et al. 2003, Rey et al. 2006, Britch et al. 2008).

In 2011, a larger and wider-distributed population than anticipated of *Ae. aegypti* was found in the city of Jacksonville, FL, during surveys conducted by the Navy Entomology Center of Excellence (NECE) and the JMCD (unpublished data). Because of the importance of both species in dengue and other arboviral transmission, this study was undertaken to further define the distribution of *Ae. aegypti* and *Ae. albopictus* in Jacksonville and to evaluate the efficacy of the Biogents™ BG-Sentinel® trap (BGS) and oviposition cups (OCs) in collecting both species.

Surveillance of mosquito species is essential to determine the risk of pathogen transmission and to monitor the impact of vector control efforts. Since even closely related mosquito species, such as *Ae. aegypti* and *Ae. albopictus*, may differ in

[1] Navy Entomology Center of Excellence, Box 43 Building 927, Naval Air Station Jacksonville, Jacksonville, FL 32212.

[2] Navy and Marine Corps Public Health Center, Center for Medical, Agricultural and Veterinary Entomology, ARS USDA, 1600 SW 23rd Drive, Gainesville, FL 32068.

[3] Jacksonville Mosquito Control Division, Jacksonville, FL 32218.

Report Documentation Page

Form Approved
OMB No. 0704-0188

1. REPORT DATE **MAR 2015**	2. REPORT TYPE	3. DATES COVERED **00-00-2015 to 00-00-2015**
4. TITLE AND SUBTITLE **Comparison of BG-Sentinel Trap and Oviposition Cups for Aedes aegypti and Aedes albopictus Surveillance in Jacksonville, Florida, USA**		5a. CONTRACT NUMBER 5b. GRANT NUMBER 5c. PROGRAM ELEMENT NUMBER
6. AUTHOR(S)		5d. PROJECT NUMBER 5e. TASK NUMBER 5f. WORK UNIT NUMBER
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) **Navy Entomology Center of Excellence,Box 43, Building 927,Naval Air Station,Jacksonville,FL,32212**		8. PERFORMING ORGANIZATION REPORT NUMBER
9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)		10. SPONSOR/MONITOR’S ACRONYM(S) 11. SPONSOR/MONITOR’S REPORT NUMBER(S)
12. DISTRIBUTION/AVAILABILITY STATEMENT **Approved for public release; distribution unlimited**		
13. SUPPLEMENTARY NOTES		
14. ABSTRACT		
15. SUBJECT TERMS		

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT **unclassified**	b. ABSTRACT **unclassified**	c. THIS PAGE **unclassified**	**Same as Report (SAR)**	**6**	

their response to a surveillance method, it is important to identify the most efficient methods and tools with which to sample the populations of interest at adequate numbers.

With locally acquired cases of dengue occurring frequently in Florida since 2010, NECE and the JMCD had great interest in determining the risk of local transmission to the citizens of Jacksonville. Accordingly, the objectives of the study were to further delineate the populations of *Ae. aegypti* in Jacksonville and to determine whether adult or egg surveillance is better for collecting both *Ae. aegypti* and *Ae. albopictus*.

MATERIALS AND METHODS

Study site

The city of Jacksonville is located in the northeastern region of Florida and has a subtropical climate with temperatures ranging from 0°C to 35°C and an average of 52 in. of rainfall per year. It is the largest city in the USA based on total area and has a population of 813,519 and an urban core located along the Saint Johns River. Ten study sites were chosen in the suburban neighborhoods of Avondale, west of the St. Johns River and east of the Jacksonville Beltway (I-295). This section of Jacksonville was chosen since several *Ae. aegypti* adults were detected in the region in 2011 during a preliminary study; thus, volunteers were solicited by the researchers in a manner as to spread the 10 locations out as much as possible within the study grid. Study sites were a mix of individual houses and churches with ornamental vegetation and a mix of hardwoods and pine trees at each site.

Surveillance

Mosquito surveillance was conducted from July 5 through September 5, 2012. Two types of traps were used: 1) The BG-Sentinel® (BGS) traps baited with 1 lb (454 g) of dry ice and Biogents human odor lures and 2) black plastic oviposition cups (OCs) lined with seed germination paper (Anchor Paper, Saint Paul, MN) and 75% full with tap water.

One BGS trap was deployed at each field site once a week at 0900 h and collected after 24 h. Four OCs were placed at each site over a 7-day period. The OCs were placed out randomly in the plot each week and were never closer than 10 ft (3.3 m) apart. Adult mosquitoes captured in the BGS traps were returned to the NECE laboratory, frozen, and identified to species. Eggs laid on the seed germination paper in the OCs were returned to the NECE laboratory, counted, identified to genus, and reared to adults for identification using identification keys of Darsie and Morris (2003). Egg papers were kept moist and then placed directly into larval rearing trays filled with dechlorinated water and held at 27°C. Larvae were fed TetraMin (Blacksburg, VA) fish food ground into a fine powder daily. Voucher specimens of each species are stored in the NECE Arthropod Collection at Naval Air Station Jacksonville and will be made available for additional study upon request. Weekly rainfall was monitored at each site using a rain gauge placed 3 ft (1 m) from the BGS. During the study, the JMCD carried out 33 ground ultralow-volume (ULV) insecticide applications in our study areas to control West Nile virus vectors. The insecticide applied was Permanone® (Montvale, NJ) 30-30, using either a London Fog© (London Fog, Long Lake, MN) or Grizzly© (Clarke, St. Charles, IL) ULV sprayers at a rate of 3.5 oz/min.

Statistical methods

Aedes aegypti *vs.* Aedes albopictus: Preliminary assessment of normality and homogeneity of variances of the distribution of mosquito counts (adults in the BGS traps, eggs in the OCs) using numerous goodness-of-fit (GOF) tests including the Kolmogorov-Smirnov test for normality and the Bartlett test for homogeneity (Zar 1999) indicated nonnormal, non-homoscedastic behavior, even after log-transformation of the data. Hence, the nonparametric Kruskal-Wallis (K-W) hypothesis test was used in lieu of the parametric analysis of variance (ANOVA) test at the 95% confidence level, for the data analysis.

One-way K-W tests were applied to abundances of *Ae. aegypti* and *Ae. albopictus* collected in BGS traps and OCs (Intel Visual Fortran Composer XE 2013, Intel Corporation, Santa Clara, CA), in which the null hypothesis (Ho) states that, at the 95% confidence level ($a = 0.05$), there is no significant difference in abundances between the BGS traps and OCs.

Rainfall and insecticide applications: Because the study site was in an area with West Nile virus fatalities in previous years, JMCD was required to carry out vector control operations that included insecticide applications using ULV truck-mounted sprayers. To determine the potential impact rainfall and vector control operations had on the abundance of *Ae. aegypti* and *Ae. albopictus* individuals collected in BGS and OCs, the dates and locations of ULV insecticide applications within the experimental area were used to define a "days after spray" variable. Because of the importance of rainfall on mosquito populations, a multiple linear regression analysis (MLRA) was conducted to assess magnitudinal and directional impacts of rainfall and time delay of spray application of insecticide on mosquito abundances (counts). For each of the 10 sites, abundances or counts (y) of *Ae. aegypti* and *Ae. albopictus*

Table 1. Mosquito species composition, abundance, and relative percent captured with BG-Sentinel™ (BGS) traps baited with Biogents human odor lures from July 5 to September 5, 2012, Jacksonville, FL.

Species	No. adults collected	% total
Aedes aegypti	5,544	38.8
Ae. infirmatus	4,401	30.8
Psorophora ferox	1,466	10.2
Ae. albopictus	1,205	8.42
Culex nigripalpus	1,027	7.18
Cx. quinquefasciatus	179	1.25
Ps. columbiae	145	1.01
Ae. vexans	123	0.86
Ae. canadiensis	58	0.40
Ae. taeniorhynchus	43	0.30
Ps. howardii	25	0.17
Cx. tarsalis	22	0.15
Ae. triseriatus	17	0.12
Anopheles crucians	16	0.11
Ps. ciliata	14	0.10
An. quadrimaculatus	6	0.04
Cx. erraticus	4	0.03
Ae. fulvus pallens	4	0.03
Toxorhynchites sp.	3	0.02
Uranotaenia sp.	1	0.01
Total	14,303	100

collected from the BGS traps ($n = 10$ collection dates) and from the OCs ($n = 8$ collection dates) were regressed versus 2 covariates: rainfall (x_1, in.) and days after spray (x_2, days), according to the regression equation: $y = a + b_1 \times x_1 + b_2 \times x_2$. Magnitude and sign of the regression coefficients for rainfall (b_1) and days after spray (b_2) were obtained to assess the degrees of sensitivity and directional impacts (e.g., positive versus negative correlation) of these covariates on adult/egg abundances. The difference in collection days was due to the loss of 2 OC samples (August 23 and August 28) attributed to incubator malfunction. In addition to the MLRA, a comparison of regressions analysis was conducted at the 95% confidence level ($a = 0.05$), to test for differences in regression slopes, elevations (i.e., regression intercepts), and coincidental regression among the N regressions. In these tests, the null hypothesis (Ho) formally states no difference in regression slopes, no difference in elevations (regression intercepts), and coincidence of the regression equations (Zar 1999).

RESULTS

Species composition

In total, 14,303 mosquitoes representing 20 species from 7 genera were collected in the BGS traps from all sites during the 10-wk study (Table 1). *Aedes aegypti* was the dominant species, followed by *Aedes infirmatus* (Dyar and Knab), *Psorophora ferox* (von Humboldt), and *Ae. albopictus*. Of the 20 species collected, 13 are known vectors, demonstrating the utility of the BGS traps for surveillance of other non-dengue vector species.

In total, 8,902 eggs, including eggs that were already hatched, were collected in the OCs, and 2,904 adults were reared. Only 2 species were reared from eggs collected in the OCs: *Ae. aegypti* and *Ae. albopictus*. There was variability across the 10 study sites, with *Ae. aegypti* being the predominate species at 7 of the 10 sites.

In the BGS traps, adult counts of *Ae. aegypti* were significantly higher than those of *Ae. albopictus* across the 10 sites ($\chi^2 = 13.17$, df $= 1$, $P < 0.0001$), suggesting that the BGS trap was more effective in collecting *Ae. aegypti* than *Ae. albopictus*, although there was proportional variation across sample sites (Table 2).

A significant difference in the total abundance (i.e., summed over the 10 sites, $n = 10$ collection

Table 2. Number of *Ae. aegypti* and *Ae. albopictus* individuals captured and percent (in parentheses) of total captured using BG-Sentinel™ traps (BGS) baited with Biogents human odor lures and in oviposition cups (OCs), with total rainfall from July 5 to September 5, 2012, at 10 sites in Jacksonville, FL.

	Aedes aegypti		*Ae. albopictus*		
Site number	No. BGS (%)	No. OCs (%)	No. BGS (%)	No. OCs (%)	Rainfall (in.)
1	810 (98.4)	109 (95.6)	13 (1.6)	5 (4.4)	7.2500
2	1,563 (91.5)	477 (93.5)	146 (8.5)	33 (6.5)	6.6
3	438 (97.5)	141 (87.04)	11 (2.5)	21 (12.96)	10.6
4	1,006 (95.9)	104 (79.4)	43 (4.1)	27 (20.6)	13.1
5	137 (33.3)	45 (11.0)	275 (66.7)	363 (89.0)	7.9
6	59 (42.7)	9 (7.9)	79 (57.3)	105 (92.1)	9.1
7	691 (57.2)	363 (49.5)	517 (42.8)	370 (50.5)	7.5
8	361 (96.0)	296 (97.4)	15 (4.0)	8 (2.6)	11.2
9	540 (95.9)	136 (70.8)	23 (4.1)	56 (29.2)	3.7
10	272 (69.9)	22 (9.3)	117 (30.1)	214 (90.7)	5.1
Total	1,586 (82.6)	1,702 (58.6)	1,240 (17.4)	1,202 (41.4)	82.4
Mean	588.65 (77.8)	170.2 (60.2)	123.96 (22.1)	120.20 (39.9)	8.2
SE ±	143.16 (0.07)	49.54 (0.11)	50.92 (0.07)	45.52 (0.11)	0.9

Table 3. Multiple linear regression analyses for total mosquito abundance at 10 sites related to rainfall and number of days after an insecticide application (DAS), $y = a + b_1 \times x_1 + b_2 \times x_2$, where y = abundance, x_1 = rainfall (in.), and x_2 = days after spray. Mosquitoes were captured using BG-Sentinel™ traps baited with Biogents human odor lures and in oviposition cups, and total rainfall was measured at each site from July 5 through September 5, 2012, in Jacksonville, FL.

Site	Equation	n	df	R^2
1	$y = 131.4356 + 10.4538 \times \text{Rain} - 2.1963 \times \text{DAS}$	9	6	0.1976
2	$y = 223.9090 + 45.0302 \times \text{Rain} - 6.7055 \times \text{DAS}$	9	6	0.4211
3	$y = 95.9966 - 59.3836 \times \text{Rain} - 6.6130 \times \text{DAS}$	9	6	0.6027
4	$y = 201.1043 + 0.0000 \times \text{Rain} - 8.1295 \times \text{DAS}$	9	6	0.2278
5	$y = 230.0070 + 141.8893 \times \text{Rain} - 106.6302 \times \text{DAS}$	9	6	0.7116
6	$y = 35.0362 + 33.4608 \times \text{Rain} - 2.6531 \times \text{DAS}$	9	6	0.3748
7	$y = 159.6640 + 23.7380 \times \text{Rain} - 8.6358 \times \text{DAS}$	9	6	0.2518
8	$y = 113.1827 - 27.9539 \times \text{Rain} - 0.5218 \times \text{DAS}$	9	6	0.3339
9	$y = 69.9945 - 0.0036 \times \text{Rain} - 0.7680 \times \text{DAS}$	9	6	0.1335
10	$y = 58.1294 - 60.3950 \times \text{Rain} + 17.8490 \times \text{DAS}$	9	6	0.9649
Total	$y = 141.3794 + 0.0011 \times \text{Rain} - 1.3320 \times \text{DAS}$	83	80	0.0152

dates) was observed between the numbers of *Ae. aegypti* (n = 5,544, mean = 587.65 adults, standard error [SE] = 132.52 adults) and *Ae. albopictus* (n = 1,205, mean = 123.96 adults, SE = 22.17 adults) captured in BGS traps (χ^2 = 13.16, df = 1 , $P < 0.01$). However, there was no significant difference in the total abundance (summed over the 10 sites, n = 8 collection dates) between *Ae. aegypti* (n = 1,702, mean = 212.75 eggs, SE = 59.75 eggs) and *Ae. albopictus* (n = 1,202, mean = 150.25 eggs, SE = 60.79 eggs) using the OCs (χ^2 = 1.46, df = 1, P = 0.23). Our study did not find a significant difference in the numbers of *Ae. aegypti* and *Ae. albopictus* in the OCs, even in areas where the BGS traps captured significantly more *Ae. aegypti* adults than *Ae. albopictus.*

Rainfall and insecticide applications: The amount of rainfall varied between sites (Table 2). The relationship between total mosquito abundance and rainfall also varied, with sites 3, 8, 9, and 10 showing a negative correlation between total mosquito abundance and rainfall (Table 3). Although when all sites were combined, rainfall had a positive correlation with total mosquito abundance, where $y = 141.379 + 0.001 \times \text{Rain} - 1.332 \times \text{Days After Spray}$, $R^2 = 0.015$ (Table 3).

The total mosquito abundance was negatively correlated with insecticide applications when all sites were combined, where $y = 141.379 + 0.001 \times \text{Rain} - 1.332 \times \text{Days After Spray}$, $R^2 = 0.0152$ (Table 3). When analyzed by site, only site 10 showed a positive correlation with abundance and days after spray ($y = 58.129 - 60.395 \times \text{Rain} + 17.849 \times \text{Days After Spray}$, $R^2 = 0.9649$). The *Ae. aegypti* and *Ae. albopictus* regressions showed negative correlations with rainfall while also showing negative correlations in abundance with days after spray (Table 4).

Comparing the regression equations for *Ae. aegypti*, *Ae. albopictus*, and both species combined, there was no difference in the regression coefficients (slopes) for rainfall (b_1) or for days after spray (b_2) ($F_{2, 240} = 0.202$, $P = 0.44$). However, there was a significant difference in elevation ($F_{2, 244} = 40.91$, $P < 0.0001$), and the regression equations were not coincident, indicating a significant difference among these 3 regressions ($F_{6, 240} = 13.505$, $F_{crit} = 2.136$, $P < 0.0001$).

DISCUSSION

Prior to our study, *Ae. aegypti* was assumed to be nearly extirpated by *Ae. albopictus* from Jacksonville, FL, with individuals being found only sporadically in the inner urban areas of the city (JMCD, unpublished data). Our findings suggest that not only is *Ae. aegypti* present, but it is the predominate *Aedes* species at the majority of sites sampled in western Jacksonville. Our results show that there were widespread populations of *Ae. aegypti* that have not been displaced by *Ae. albopictus*, despite both species coexisting at each site.

Table 4. Multiple linear regression analysis results for *Ae. aegypti* and *Ae. albopictus* related to rainfall and number of days after an insecticide application (DAS), $y = a + b_1 \times x_1 + b_2 \times x_2$, where y = abundance, x_1 = rainfall (in), and x_2 = days after spray. Mosquitoes were captured using BG-Sentinel™ traps baited with Biogents human odor lures and in oviposition cups, and total rainfall was measured at each site from July 5 through September 5, 2012, in Jacksonville, FL.

Species	Equation	n	df	R^2
Ae. aegypti	$y = 56.0525 - 0.0021 \times \text{Rain} - 0.5186 \times \text{DAS}$	83	80	0.0134
Ae. albopictus	$y = 13.2872 - 0.00001 \times \text{Rain} - 0.2480 \times \text{DAS}$	83	80	0.0244

Our study also demonstrated that the OCs results indicated similar abundances of *Ae. aegypti* and *Ae. albopictus*, whereas the BGS results suggested higher levels of *Ae. aegypti* adults in those same areas. The more anthropophilic behavior of *Ae. aegypti* may have led to an increased attraction to the BGS trap due to the human-chemical–derived lure used with the traps. Since the lure is not used in the OCs, this could explain the lack of difference in attraction if both species are equally attracted to the visual cues of the white and black color of the traps. The more zoophilic nature of *Ae. albopictus* may make it less attracted to the BGS traps baited with the lure. The BGS trap and lure were specifically designed to collect *Ae. aegypti*, and reliance on just the BGS traps alone may not accurately reflect the true ratios of *Ae. aegypti* and *Ae. albopictus* when these 2 species coexist (Barrera et al. 2011, Crepeau et al. 2013) .

Rainfall was found to be positively correlated with mosquito abundance across all sites, but it was negatively correlated with the numbers of *Ae. aegypti* and *Ae. albopictus*. The amount of rain is a known environmental factor that influences the populations of mosquitoes, including *Ae. aegypti* and *Ae. albopictus* (Barrera et al. 2011, Reiskind and Lounibos 2012). In suburban communities, such as Avondale, home irrigation of yards and ornamental plantings may also play an important role in maintaining stable populations of both species during low rainfall periods, decreasing the effect of seasonal rains (Reiskind and Lounibos 2012). Both trap types are suitable for measuring the impact of rainfall on the population numbers of *Ae. aegypti* and *Ae. albopictus*.

Abundance of mosquitoes in the BGS traps and OCs measured the impact of the control efforts carried out by the JMCD against West Nile virus vector species. Both trapping methods detected the impact of the ground ULV sprays at 9 of 10 sites, but we are unsure if the insecticide applications lowered the *Aedes* populations below a threshold necessary to prevent dengue virus transmission. Even small populations of *Ae. aegypti* have been determined to maintain virus transmission, so other vector control methods (e.g., source reduction) would have to be conducted simultaneously with ULV application of insecticides to stop the transmission (WHO 2009, Esu et al. 2010, Stoddard et al. 2013).

Due to the ongoing risk of West Nile virus in Jacksonville, we were unable to establish an untreated control area to accurately estimate the impact of ULV spraying on the *Aedes* populations. It may be that in untreated areas, populations of *Ae. aegypti* would have been even higher.

Our results show that in order to accurately determine vectorborne disease threats and impact of control operations on these 2 species, multiple trapping techniques should be considered when studying population dynamics between *Ae. aegypti* and *Ae. albopictus*. Information regarding sensitivity of OCs and BGS traps for the detection of critical disease vectors is instrumental to surveillance and control programs where both *Ae. aegypti* and *Ae. albopictus* coexist. As more and more literature suggests that *Ae. aegypti* populations are expanding, and resident *Ae. albopictus* populations are being displaced, the ability to accurately determine species composition will become instrumental in the control of arthropodborne pathogens (O'Meara et al. 1993, Sibal 1994, Moore and Mitchell 1997).

ACKNOWLEDGMENTS

We thank Jason Francona, Michael Denson, Dante Benedicto, Gregory Allen, and Marcus McDonough for assistance with field collections. The views expressed in this article are those of the authors and do not necessarily reflect the official policy or position of the Department of the Navy, Department of Defense, nor the US government. This work was funded by the Deployed War Fighter Program. The author and coauthors are employees of the US government. This work was prepared as part of our official duties. Title 17 U.S.C. section 17 provides that, "Copyright protection under this title is not available for any work of the United States government." Title 17 U.S.C. section 101 defines a US government work as a work prepared by a military service member or employee of the US government as part of that person's official duties.

REFERENCES CITED

Bargielowski I, Lounibos LP. 2014. Rapid evolution of reduced receptivity to interspecific mating in the dengue vector *Aedes aegypti* in response to satyrization by invasive *Aedes albopictus*. *Evol Ecol* 28:193–203.

Barrera R. 1996. Competition and resistance to starvation in larvae of container inhabiting *Aedes* mosquitoes. *Eco Entomol* 21:117–127.

Barrera R, Amador M, MacKay AJ. 2011. Population dynamics of *Aedes aegypti* and dengue as influenced by weather and human behavior in San Juan, Puerto Rico. *PLoS Negl Trop Dis* 5(12):e1378. doi:10.1371/journal.pntd.0001378.

Bhatt S, Gething PW, Brady OJ, Messina JP, Farlow AW, Moyes CL, Drake JM, Brownstein JS, Hoen AG, Sankoh O, Myers MF, George DB, Jaenisch T, William Wint GR, Simmons CP, Scott TW, Farrar JJ, Hay SI. 2013. The global distribution and burden of dengue. *Nature* 495:504–507.

Braks MAH, Honorio NA, Lourenco-De-Oliveira R, Juliano SA, Lounibos LP. 2003. Convergent habitat segregation of *Aedes aegypti* and *Aedes albopictus* (Diptera: Culicidae) in southeastern Brazil and Florida. *J Med Entomol* 40:785–794.

Britch SC, Linthicum KJ, Anyamba A, Tucker CJ, Pak EW, the Mosquito Surveillance Team. 2008, Year long-term surveillance data and patterns of invasion by

Aedes albopictus in Florida. *J Am Mosq Control Assoc* 24:115–120.

Crepeau TN, Healy SP, Bartlett-Healy K, Unlu I, Farajollahi A, Fonseca DM. 2013. Effects of Biogents Sentinel trap field placement on capture rates of adult Asian tiger mosquitoes, *Aedes albopictus*. *PLoS ONE* 8(3):e6052. doi:10.1371/journal.pone.0060524.

Damal K, Murrell EG, Juliano SA, Conn JE, Loew SS. 2013. Phylogeography of *Aedes aegypti* (yellow fever mosquito) in south Florida: mtDNA evidence for human-aided dispersal. *Am J Trop Med Hyg* 29:482–488.

Darsie RF, Morris CD. 2003. Keys to the Ault Females and Fourth Instar Larvae of the Mosquitoes of Florida (Diptera, Culicidae). *Technical Bulletin of the FMCA. Vol 1(revised)*. Florida Mosquito Association, INC. Fort Meyers, FL.

Esu E, Lenhart A, Smith L, Horstick O. 2010. Effectiveness of peridomestic space spraying with insecticide on dengue transmission; systematic review. *Trop Med Int Health* 15:619–631.

Gerhardt RR, Gottfried KL, Apperson CS, Davis BC, Erwin PC, Smith AB, Panella NA, Powell EE, Nasci RS. 2001. First isolation of La Crosse virus from naturally infected *Aedes albopictus*. *Emerg Infect Dis* 7:807–811.

Ibanez-Bernal S, Briseno A, Mutebi JP, Argot E, Rodriguez G, Martinez-Campos C, Paz R, Roman PF, Tapia-Conyer R, Flisser A. 1997. First record in America of *Aedes albopictus* naturally infected with dengue virus during the 1995 outbreak at Reynosa, Mexico. *Med Vet Entomol* 4:305–309.

Moore CG, Mitchell CJ. 1997. *Aedes albopictus* in the United States: ten-year presence and public health implications. *Emerg Infect Dis* 3:329–334.

O'Meara GF, Gettman AD, Evans LF, Curtis GA. 1993. The spread of *Aedes albopictus* in Florida. *Am Entomol* 32:554–562.

O'Meara GF, Gettman AD, Evans LF, Scheel FD. 1992. Invasion of cemeteries in Florida by *Aedes albopictus*. *J Am Mosq Control Assoc* 8:1–10.

Reiskind MH, Lounibos LP. 2012. Spatial and temporal patterns of abundance of *Aedes aegypti* L. (Stegomyia aegypti) and *Aedes albopictus* (Skuse) [Stegomyia albopictus (Skuse)] in southern Florida. *Med Vet Entomol* 27:421–429.

Rey JR, Nishimura N, Wagner B, Braks MAH, O'Connell SM, Lounibos LP. 2006. Habitat segregation of mosquito arbovirus vectors in south Florida. *J Med Entomol* 43:1134–1141.

Schliessmann RL, Magennis NJ. 1964. Initial plan for the eradication of *Aedes aegypti* from the United States. *Pest Control* 7:34–38.

Sibal IH. 1994. *Aedes albopictus* and *Aedes aegypti* in Polk County. *J Fl Mosquito Control Ass* 26:15–16.

Stoddard ST, Forshey BM, Morrison AC, Paz-Soldanf VA, Vazquez-Prokopec GM, Astetec H, Reiner RC, Vilcarromero S, Elderh JP, Halsey ES, Kochel TJ, Kitron U, Scott TW. 2013. House-to-house human movement drives dengue virus transmission. *Proc National Acad Sci USA* 110:994–999.

Turell MJ, Dohm DJ, Sardelis MR, O'Guinn ML, Andreadis TG, Blow JA. 2005. An update on the potential of North American mosquitoes (Diptera: Culicidae) to transmit West Nile virus. *J Med Entomol* 42:57–62.

WHO [World Health Organization]. 2009. *Dengue, guidelines for diagnosis, treatment and control*. Geneva, Switzerland: World Health Organization.

Zar JH. 1999. *Biostatistical analysis.* Fourth edition. Upper Saddle River, NJ: Prentice Hall.