

TECHNICAL BULLETIN OF THE FLORIDA MOSQUITO CONTROL ASSOCIATION



VOLUME 9, 2013



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OF THE
FLORIDA MOSQUITO
CONTROL
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FLORIDA MOSQUITO CONTROL ASSOCIATION, INC. ORGANIZED IN 1922

The Florida Mosquito Control Association, Inc. is a non-profit, technical, scientific, and educational association of mosquito control, medical, public health, and military biologists, entomologists, engineers, and lay persons who are interested in the biology and control of mosquitoes or other arthropods of public health importance.

TECHNICAL BULLETIN OF THE FLORIDA MOSQUITO CONTROL ASSOCIATION

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The Technical Bulletin of the Florida Mosquito Control Association is published by The Florida Mosquito Control Association, Inc.

Printed by the
E. O. Painter Printing Company
P.O. Box 877
DeLeon Springs, FL 32130

ARBOVIRUS SURVEILLANCE AND MOSQUITO CONTROL WORKSHOP

A volume of selected papers from:

- The 5th workshop, March 26-28, 2008
- The 6th workshop, March 31-April 2, 2009
- The 7th workshop, March 23-25, 2010
- The 8th workshop, March 29-31, 2011
- The 9th workshop, March 27-29, 2012
- The 10th workshop, March 26-28, 2013

Edited by:
Rui-De Xue

Sponsored by:
Anastasia Mosquito Control District,
St. Johns County, St. Augustine, Florida
and
USDA-ARS, Center for Medical, Agricultural, and Veterinary Entomology,
Gainesville, Florida



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INTRODUCTION

Between 2008 and 2013 the Anastasia Mosquito Control District (AMCD) of St. Johns County, Florida held its 5th, 6th, 7th, 8th, 9th, and 10th annual Arbovirus Surveillance and Mosquito Control Workshops at District headquarters in St. Augustine. All workshops were sponsored by the AMCD and the USDA-ARS, Center for Medical, Agricultural, and Veterinary Entomology (CMAVE) in Gainesville, FL. These workshops were designed to facilitate the exchange of information regarding mosquito-borne diseases, review recent research and developments in arbovirus transmission, mosquito surveillance, mosquito management, and to offer unique training opportunities for mosquito control professionals.

The 5th workshop was held from March 26-28, 2008 and included 40 presentations. Dave Brown from Sacramento-Yolo Vector Control District, Sacramento, CA and John Edman from University of California, Davis, CA, presented the keynote addresses about West Nile virus. Scientists from India and Israel gave their presentations about mosquito-borne diseases in India and sugar bait studies in Israel. Scientists from USDA and universities gave their presentations about surveillance and control of mosquitoes and mosquito-borne diseases. Industry representatives gave their presentations about new products and equipment for the upcoming mosquito season.

The 6th workshop was held from March 31 -April 2, 2009 and included 46 presentations. Lyle Petersen from Centers for Disease Control and Prevention (CDC), Ft. Collins, CO presented the keynote address about mosquito-borne arboviral diseases in the USA. Scientists from Egypt, China, Thailand, and Israel presented talks about Rift Valley Fever, mosquito behavior and insecticide-impregnated bednet studies in Asia. There were many presentations from federal, state, universities, local mosquito control programs, and industry. During the workshop the AMCD celebrated its 60th anniversary.

The 7th workshop was held from March 23-25, 2010 and included 53 presentations covering a variety of diverse topics. Kenneth Linthicum, USDA/CMAVE, gave the keynote address about the future direction for mosquito control. Three scientists from China and one from Israel gave presentations about arthropod-borne viruses and mosquito surveillance programs in China and sugar baited mosquito control in Israel. Scientists from USDA, University of Florida, state governmental agencies, local mosquito control programs, and industry gave several presentations.

The 8th workshop was held from March 29-31, 2011 and included 54 presentations in seven sessions. Daniel Strickman, from USDA, National Program gave the key note speech about Chickungunya and the national needs for research and development. Tong-Yan Zhao from China reported about an outbreak of Chickungunya in Southern China in 2010. Err-Lieh Hsu gave a presentation about dengue fever in Taiwan. Other scientists from China (6), Israel (2), Australia (1), and Greece (1) gave presentations about dengue fever, vector surveillance, and vector control techniques. Scientists from universities, USDA, US Navy, state and local mosquito control programs, and industry made presentations.

The 9th workshop was held from March 27-29, 2012 and included 62 presentations divided into 9 sessions. Robert Wirtz from CDC, Atlanta, GA gave the keynote address about their research programs in Atlanta. Gunter Muller presented an invited talk about toxic sugar baits for mosquito and sandfly control. Scientists from China (5), Israel (2), and Greece (1) and several university, federal, state, and local mosquito control programs made presentations about malaria control, insecticide resistance monitoring, and population surveillance.

The 10th workshop was held from March 26-28, 2013, included 73 presentations divided into 7 sessions. Due to the recent resurgence of West Nile virus (WNV) in the USA

during 2012, the prevention and control of WNV was the theme for this workshop. Roger Nasci from CDC, Ft. Collins, CO gave the keynote address about WNV and other viruses in the USA. Scientists from the USDA, US Navy, universities, and mosquito control professionals from state and local governmental agencies as well as industry gave presentations that included updates of their mosquito surveillance and control programs and a review of new products and equipment. Additionally, there were many presentations about the surveillance and control of *Aedes albopictus*.

A total of 98 continuing education credits (CEU) were provided to workshop attendees between 2008 and 2013. We are especially appreciative of the speakers and contributors who gave presentations in the workshops and those who submitted manuscripts for inclusion in this volume, as well

as the organizations/companies who provided partial funding for the workshops. We thank those who reviewed the manuscripts prior to publication of this volume of the Technical Bulletin of the Florida Mosquito Control Association, including Tom Unnasch, Whitney Qualls*, John Beier*, James Becnel, Larry Hribar*, Peter Jiang*, Gunter Muller*, James Cilek*, Jonathan Day*, Sandy Allan*, Seth Britch*, Muhammad Farooq, Harry Zhong*, Abu Ahmad, Ary Farajollahi, Graham White, Aaron Lloyd, Jodi Scott, Tim Hope* and Alice Fulcher* (* indicates reviewers who reviewed more than one manuscript). We also acknowledge the support and encouragement of the AMCD Board of Commissioners, administrative office, District staff, and industry.

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THE BIOLOGY OF ARBOVIRAL TRANSMISSION IN FLORIDA

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ABSTRACT. All mosquito-borne virus transmission cycles have at least three biotic components; a viral pathogen, a mosquito vector, and a vertebrate amplification host. In addition to the biological components, these disease transmission cycles are driven by environmental (abiotic) factors that serve to synchronize viral transmission between mosquito vectors and vertebrate hosts. Understanding the biotic and abiotic effects on virus transmission provides information to forecast where and when outbreaks of arboviral transmission may occur. Surveillance strategies to monitor virus transmission must include measures of virus, mosquito, and host animal abundance, transmission rates, and mosquito population age structures as well as daily measures of environmental conditions including rainfall and temperature. A well-designed arboviral surveillance program should be used to establish long term regional baseline data that can be compared with real-time data to accurately evaluate the risk of arboviral amplification and epidemic or epizootic disease transmission.

Key Words: arboviruses, disease transmission, mosquitoes, epidemic risk

I. INTRODUCTION

Mosquito-borne viruses, or arboviruses, infect and may cause disease in humans, domestic animals, and wildlife. Several arboviruses have played an important role in Florida public health. Among the earliest known arboviruses reported in Florida were yellow fever (YFV) and dengue viruses (DENV). The first suspected outbreak of yellow fever occurred in Union sailors patrolling near Cape Canaveral in 1864 (Patterson 2004) but it wasn't until 1901 that YFV was proven

to be a mosquito-borne viral pathogen. During the period of territorial development (1821-1945) Florida coastal towns were periodically decimated by outbreaks of yellow fever. However, the largest and most devastating yellow fever epidemics were reported in Key West (nine outbreaks between 1867 and 1888), Jacksonville (1877 and 1888), and Pensacola (1905). Florida's first documented dengue epidemic was reported in Pensacola during the summer of 1827 after which DENV transmission was not reported again in Florida until 1898 (Day 2008). However, other outbreaks believed to have been dengue occurred in Florida beginning in the mid-1800s. Significant dengue epidemics were reported during 1921 and 1922 in Jacksonville, Tampa, and St. Petersburg. The most extensive Florida dengue epidemic was focused in Miami during the summer of 1934 and involved an estimated 25,000 human cases that were spread throughout the Florida peninsula (Ehrenkranz et al. 1971).

More recently, three mosquito-borne encephalitis viruses have played an important role in Florida public health. Saint Louis encephalitis virus (SLEV) has been responsible for five Florida epidemics (1959, 1961, 1962, 1977, and 1990) all of which were centered in the Florida peninsula. The SLEV was first reported from Florida in association with a focal outbreak in Miami during the summer of 1952. Since 1952, there have been 736 human SLE cases reported in Florida. The peak transmission period for SLEV to humans occurs during August, September, and October.

The first two human cases of eastern equine encephalitis virus (EEEV) were reported in Florida during the summer of 1957. Since 1957, there have been 78 human eastern equine encephalitis cases reported

in the state. The peak transmission months for EEEV in Florida are June through August. Horses are frequently infected with EEEV. Since 1982, a total of 2,179 EEEV-positive horses (for an annual average of 70.3) have been reported in Florida.

In August of 2001, West Nile virus (WNV) was first reported in Florida. Since that time 290 human cases of West Nile have been reported throughout the state. The peak transmission months for WNV in Florida are July through September. Horses are frequently infected with WNV. Since the summer of 2001, a total of 1,175 WNV-positive horses (for an average of 107 per year) have been reported in Florida.

Arboviral epidemics can result in significant health and economic consequences (Day and Stark 2000). Epizootics of EEEV and WNV in horses result in losses of livestock and expenses to horse owners. Significant financial impacts related to public health, the economy, and tourism have been reported for outbreaks caused by EEEV (Vilari et al. 1995), SLEV (Harden et al. 1991), and WNV (Zohrabian et al. 2004). As is the case with vector-borne disease transmission in other parts of the world, arboviruses in Florida have reduced the health and well-being of humans, domestic animals, and wildlife throughout the state.

All mosquito-borne disease cycles involve the interplay between at least three biotic entities. These are the virus, the mosquito vector, and a variety of vertebrate host species. The mosquito-borne disease cycle is facilitated and enhanced by environmental conditions that favor the successful reproduction and propagation of the arbovirus. These environmental factors include, but are not limited to, rainfall, ground water levels, and daily high and low temperatures (Day 2001). Efficient arboviral transmission depends on an abundance of infective mosquito vectors. Most of the time, habitats contain few, if any, infective mosquitoes. A fine balance exists between the environmental conditions that support efficient viral amplification and the conditions that completely shut down arboviral transmission. Without the environmental conditions that produce a "perfect storm" of

arboviral transmission, the realization of an epidemic is unlikely. The "perfect storm" for an arboviral epidemic results in efficient cycling of virus between mosquito vectors and amplification vertebrate hosts in a manner that produces high numbers of infectious mosquitoes. All arbovirus epidemics require the following *primers*: an abundance of virus, vectors, susceptible vertebrate hosts, an aging vector population, and the susceptibility of vectors and amplification hosts to viral infection. In addition, all epidemics have a series of *triggers* that include: environmental conditions that allow the independent biological components of epidemic transmission (virus, vector, vertebrate hosts) to cycle in a way that maximizes the probability of a large number of infectious vectors and an increased probability of epidemic transmission.

It is possible to track and forecast viral amplification and to predict the risk of epidemic transmission by monitoring the presence of virus, vector abundance, vector population age structure, vertebrate host abundance, vertebrate host age, and vertebrate host susceptibility to infection. The purpose of this paper is to review what is currently known about the biology of several important arboviruses in Florida and to outline how knowledge of the biotic and abiotic components of each arboviral transmission cycle can be used to forecast and predict epidemics.

II. ARBOVIRUS BIOLOGY

Arboviral transmission occurs when viruses are present in an area year round or when they are reintroduced into an area where amplification and subsequent viral transmission is supported. The arboviruses EEEV, SLEV, and WNV are endemic in Florida. Sentinel chicken surveillance programs are maintained in many Florida counties by public health units or by mosquito control districts. Some counties maintain sentinel flocks continually and there are sentinel chicken seroconversion records that indicate that arboviral transmission occurs in some areas of Florida during every month

of the year (Day and Lewis 1992). Continuous low-level viral transmission is most likely to occur south of the east-west line between Vero Beach and Tampa, Florida (at approximately latitude 27° 35') above which the Florida climate and ecology is temperate and below which it is subtropical. In north Florida and in the Florida panhandle severe winter cold may limit year round arboviral transmission to the years where mild winters are reported.

In temperate regions north of Florida where winter freezes curtail insect activity, arboviruses are forced to either overwinter or to be periodically reintroduced into the area. One of the most likely ways that viruses overwinter is in infected adult mosquitoes that survive in underground or protected hibernacula. Mosquitoes in the genus *Culex*, which represent the primary vectors of SLEV and WNV, overwinter as adult females. Overwintering *Culex pipiens* Say females infected with SLEV were collected in Maryland and Pennsylvania during 1977 (Bailey et al. 1978). Likewise, overwintering *Cx. pipiens* females infected with WNV were collected in New York (Nasci et al. 2001) and New Jersey (Farajollahi et al. 2005). *Culex* females are long-lived (Day and Curtis 1994) and it is possible that infected adult females can survive in underground habitats such as gopher tortoise burrows or culverts during long periods of drought in Florida.

Arboviruses can also overwinter in hibernating ectothermic animals like reptiles and amphibians that have long term viremias. Laboratory experiments have shown that hibernating garter snakes infected with western equine encephalitis virus (WEEV) infected blood fed *Cx. tarsalis* Coquillett the following spring (Thomas and Eklund 1962). Similar observations were made by White et al. (2011) in laboratory experiments with EEEV-infected garter snakes in Florida. Serum samples from *Agkistrodon contortrix* and *Agkistrodon piscivorus*, the copperhead and cottonmouth vipers, respectively, were determined to be RNA positive for EEEV. These data support the possibility that these two viper species can serve as overwintering hosts (Bingham et al. 2012). In addition to snakes,

turtles, tortoises (Bowen 1977), and alligators (Klenk et al. 2004) have been proposed as species that may have long term viremias that would allow arboviruses to overwinter in hibernating animals and reemerge the following spring when competent mosquito vectors take their first blood meals.

Arboviruses can also be reintroduced into habitats that contain competent mosquito vectors and vertebrate hosts that can amplify the reintroduced virus. Long periods have typically occurred between dengue outbreaks in Florida. Florida port cities were at risk of dengue reintroduction and resulting dengue epidemics from 1827 through 1946. The source of DENV for the first documented Florida dengue epidemic in 1827 in Pensacola was an ongoing epidemic in the Virgin Islands that spread throughout the Caribbean into northern South America and into the southern USA including the port cities of Charleston, Savannah, New Orleans, and Pensacola (Patterson 2004). Following the 1827 Pensacola epidemic, DENV virtually disappeared from Florida until 1898. Rerick (1902) suggested that DENV was reintroduced into Florida in 1898 due to troop movement between Florida and Cuba in 1897 during the Spanish-American War when a significant dengue outbreak was reported in Havana. Dengue virus was again reintroduced into Jacksonville, Tampa, and St. Petersburg in 1921. This reintroduction marked the first time that DENV transmission was sustained in Florida cities during successive years. Summer and autumn transmission events were observed in 1921 and 1922 in many major urban centers of Florida (Patterson 2004). The largest dengue epidemic reported in Florida began in Miami during mid-July, 1934 (Griffitts 1935). Ehrenkranz et al. (1971) have suggested that the source of the epidemic was transport of DENV-infected U.S. troops from Haiti to Key West during the U. S. intervention in Haiti, and then on to Miami for recovery. Between July 16 and November 24, 1934 the epidemic spread from Miami north through the Florida peninsula, into North Florida and the Florida panhandle and, eventually, into southern Georgia (Ehrenkranz et al. 1971).

The most recent reintroduction of DENV into Florida occurred in Key West in 2009 (Radke et al. 2012). The virus was likely introduced from South America and transmission persisted through the 2009 and 2010 summer transmission seasons after which the virus again may have disappeared from Key West. However, sporadic introductions of DENV into south Florida continued to be reported during 2010-2012.

It is possible that the mosquito-borne encephalitis viruses (EEEV, SLEV, and WNV) are periodically reintroduced into Florida. A likely source of this reintroduction is infected migrating birds. The spring (south to north) migration may reintroduce EEEV or WNV through infected birds moving from overwintering sites in the Caribbean Islands or Central and South America. Likewise, north to south avian migration patterns in the autumn may reintroduce viral strains from the northern avian breeding sites. Since there are no likely non-primate hosts for YFV and DENV it is likely these viruses arrived in Florida via infected mosquitoes or humans at various times. However, evidence for the reintroduction of the encephalitis viruses into Florida is not as convincing as the evidence in support of the periodic reintroduction of DENV or YFV into the state.

III. MOSQUITO BIOLOGY

The primary epidemic and epizootic mosquito vectors for many of the mosquito-borne arboviral diseases in Florida are well understood. For example, *Aedes aegypti* (L.) has long been known to be the primary vector of DENV and YFV in Florida and throughout the world (Gubler 1998). During recent years, however, dengue outbreaks involving *Ae. albopictus* (Skuse) as the primary epidemic vector have emerged around the globe. A dengue outbreak was reported in Hawaii during 2001 and 2002 where *Ae. albopictus* was the primary epidemic vector (Effler et al. 2005). Likewise, several *Ae. albopictus*-driven dengue epidemics have been reported in China (Xu et al. 2007). These localized changes in the primary epidemic dengue vector have much to do with the recent his-

tory of *Ae. albopictus* dispersal and the ability of this species to invade new habitats where they quickly become firmly established (Rezza 2012).

Transmission of the mosquito-borne encephalitis viruses in the family Flaviviridae is clearly linked to mosquitoes in the genus *Culex*. This is true in the U.S. as well as world-wide (Hayes 1988). The transmission of SLEV and WNV in North America is linked to the abundance and reproductive biology of *Culex* species found in different regions of the continent. In the northern half of the USA and southern Canada the primary epidemic vector is *Cx. pipiens pipiens*. In the southern half of the country, with the exception of Florida, the primary epidemic vector is *Cx. pipiens quinquefasciatus* Say. In the western states *Cx. p. quinquefasciatus* and *Cx. tarsalis* are the primary SLEV and WNV epidemic vectors. In Florida the primary epidemic flavivirus vector is *Cx. nigripalpus*.

The epizootic transmission of EEEV is well understood and involves the amplification of EEEV between wild birds and *Culiseta melanura* (Coquillett) (Weaver et al. 1991). *Culiseta melanura* shows a strong preference for birds and is clearly not the epidemic vector that transmits EEEV to humans, nor is it the epizootic vector that transmits EEEV to horses. The mosquito species that provide a "bridge" for EEEV from infected birds to humans and horses are not well understood. However, based on viral isolations from field-collected female mosquitoes (Weaver et al. 1991) and mosquito blood feeding behavior (Cohen et al. 2009) they likely include *Cx. nigripalpus* Theobald, spring floodwater *Aedes*, *Mansonia* spp., and *Coquillettidia* spp.

Many different mosquito species can acquire and transmit arboviruses, but few are efficient epidemic and epizootic vectors. What makes an excellent arboviral vector? In general, excellent vectors are abundant, widespread (temporally and spatially), and are opportunistic blood feeders that are likely to seek and blood feed on hosts in a variety of habitats (Day 2005). Some species, such as *Wyeomyia vanduzeei* Dyar and Knab, are focally distributed in habitats that are largely determined by the presence of suit-

able oviposition sites. Females of this species may occasionally blood feed on an infected bird that enters the mosquito's habitat and acquire an arboviral infection. It is unlikely that infected *Wyeomyia* mosquitoes will disperse and carry the virus into adjacent habitats because this species tends to remain within a very limited home range.

An example of a superb arboviral vector in Florida is *Cx. nigripalpus*. Of the approximately 80 mosquito species reported in the state, *Cx. nigripalpus* alone serves as the primary epidemic vector of SLEV and WNV, the primary epizootic vector of WNV, and is likely the primary epidemic and epizootic EEEV vector (Weaver et al. 1991, Cohen et al. 2009). *Culex nigripalpus* possesses all of the traits of an excellent arboviral vector. It is abundant in most Florida counties. Because it is a subtropical species, it is closely tied to the Florida wet season from May through October when it becomes one of the most widespread and abundant mosquito species reported in the state (Day and Curtis 1989). It is found in a variety of habitats ranging from urban parks and neighborhoods to the most rural habitats in the state. It is an opportunistic blood feeder with a preference for wild birds (Edman 1974, Mackay et al. 2010). Most importantly, it shifts blood feeding from preferring avian hosts in the spring and early summer (when arboviral amplification is most prevalent) to preferring mammals in the late summer and fall (when epidemic transmission is most common) (Edman and Taylor 1968). A final factor that makes *Cx. nigripalpus* an excellent arboviral vector, as will be explained below, is that this species is profoundly affected by rainfall patterns (Day and Curtis 1994).

In summary, excellent arboviral vectors share some common traits. They are susceptible to viral infection and do not possess barriers that inhibit salivary gland infections. They are common and widespread over time and space. They are opportunistic blood feeders with a preference for vertebrate hosts that are important in the amplification of the specific arboviruses that the vector transmits. They are influenced

by environmental factors that synchronize vector and amplification host behaviors in a way that maximizes amplification efficiency and results in large numbers of infective mosquitoes resulting in a high probability of epidemic and/or epizootic transmission.

IV. AMPLIFICATION HOST BIOLOGY

Amplification (Figure 1) of the arbovirus is the key event that defines epidemic and epizootic arboviral transmission. Amplification hosts are the vertebrates that produce viremias sufficient to maximize the number of infectious mosquitoes in the habitat where amplification occurs. For example, humans serve as amplification hosts for DENV. Susceptible *Ae. aegypti* females feed on a viremic human host, acquire and incubate the virus, develop a salivary gland infection thus becoming infectious, and transmit the DENV to additional susceptible human hosts. This is an example of a relatively simple arboviral amplification cycle.

Wild birds serve as the primary amplification hosts for the mosquito-borne encephalitis viruses in Florida. Similar complexes of avian species serve as amplification hosts for both SLEV and WNV in the state (Day and Stark 1999). In the case of EEEV, avian species associated with hardwood freshwater swamps serve as the amplification hosts for the enzootic portion of the EEEV amplification and transmission cycles. A different suite of avian species serve as amplification hosts in the secondary amplification foci that become established outside of the swamps during years when EEEV expands into habitats that are not associated with the primary amplification foci (Crans et al. 1994).

The factors that make an excellent amplification host are similar to those that make an excellent vector. Amplification hosts must be susceptible to viral infection and must circulate enough virions to infect substantial numbers of blood feeding mosquitoes. Because the vertebrate host immune system usually clears an arboviral infection, the longer virus circulates at levels sufficient to infect vector mosquitoes, the more efficient the amplification cycle. Susceptible amplification hosts

Arboviral Amplification

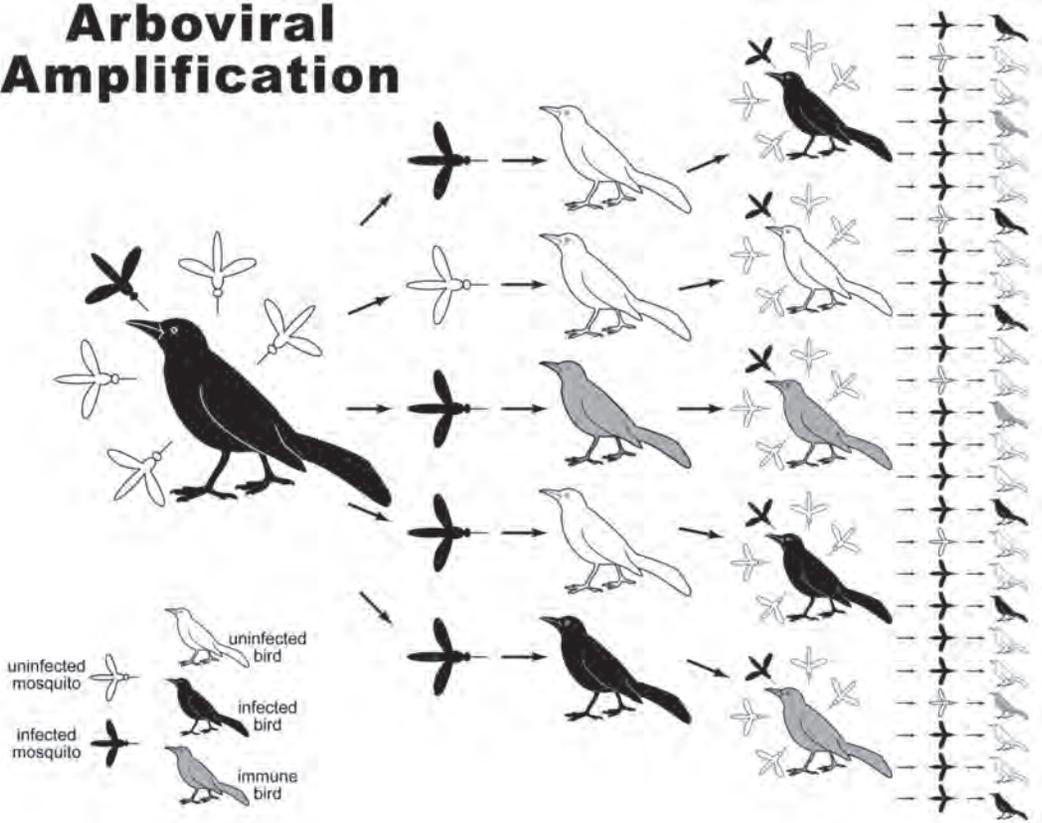


Figure 1. Arboviral amplification.

must be abundant as well as spatially and temporally widespread. As is the case with mosquito vectors, many avian species are susceptible to arboviral infection, but few serve as efficient amplification hosts. Rare avian species may become infected with virus, but are unlikely to be efficient amplification hosts due to their low number of encounters with mosquito vectors. Amplification hosts, such as turkey vultures, that are spatially clustered, may serve as focal amplifiers of virus, but do not play an important role in widespread epidemic amplification because of the focal nature of their nighttime roosting behavior. Avian species that are susceptible to viral infection, abundant, widespread (spatially and temporally), and likely to encounter host seeking vector mosquitoes are most likely to serve as primary epidemic amplification hosts (Day and Stark 1999).

The sequence of mosquito-borne encephalitis amplification is shown in Figure 1.

The bird at the left represents an infected individual that is being fed on by one infectious and four susceptible mosquitoes. Once they acquire virus, the susceptible mosquitoes undergo a temperature-dependent incubation period at the conclusion of which some will be infectious. They, along with uninfected mosquitoes in the population, feed on additional birds; some of which are susceptible to the virus and some of which are immune due to prior infection. The same sequence of events is repeated multiple times resulting in the large number of infectious mosquitoes shown on the right side of Figure 1. A large number of infectious mosquitoes increase the probability that infectious mosquitoes will encounter susceptible humans, and horses in the case of WNV and EEEV, and initiate epidemic and epizootic transmission. Amplification can, and often does, break down in several ways. Too few mosquitoes or susceptible hosts will stop am-

plification. For example, too many immune vertebrates will reduce the numbers of susceptible hosts and may stop amplification. An absence of mosquito oviposition sites will stop amplification by reducing the numbers of host seeking mosquitoes (Day and Curtis 1994). Efficient amplification depends on the presence of virus and the production of high numbers of susceptible vectors and amplification hosts that come into focused contact at a time and place that initiates the cascade of virus transmission between vertebrates and vectors depicted in Figure 1.

V. ENVIRONMENTAL BIOLOGY

The three biotic factors (virus, vector, and vertebrate host) that make up arboviral transmission cycles are independently influenced by environmental conditions, especially rainfall, drought, and temperature. However, periodically these same environmental factors cause the three biotic factors to synchronize resulting in arbovirus epidemics (Day 2001). The natural cycling of the virus depends on a reliable supply of susceptible vectors and susceptible amplification hosts. If either of these disappears, the virus may be driven to extinction. This is likely what happened in Key West, Florida following the introduction of DENV Type 1 in 2009. The virus was transmitted in Key West by *Ae. aegypti* where there was a sufficient number of susceptible humans to allow transmission to persist for two successive transmission seasons. Human cases of dengue were not reported in Key West after the 2010 transmission season, possibly due to a reduction in the number of susceptible humans, a change in the *Ae. aegypti* population, vector abundance, failure of DENV reintroduction and/or some combination of these possibilities.

The natural cycling of *Cx. nigripalpus* populations in south Florida depends almost entirely on annual rainfall patterns. During years of severe drought the mosquito is difficult to find but during years of intermittent heavy rainfall the species prospers (Day and Curtis 1993). Likewise, the natural cycling of avian amplification

hosts associated with the transmission of SLEV and WNV in peninsular Florida depends on rainfall patterns during the avian nesting season (April, May, and June) (Day and Stark 1999) as well as winter freezes in the Florida subtropical epidemic zone (Day 2001, Day and Shaman 2009).

During most years in Florida, the biological cycles of the virus, vector, and amplification host are completely separate events, each with little impact on the other. During some years it is difficult to document arbovirus transmission in Florida. However, during some years environmental factors, especially rainfall and winter freezes, coordinate the independent biological cycles and allow them to synchronize and maximize arboviral amplification and transmission. Environmental conditions favored the amplification and transmission of SLEV in Florida in 1977 and again in 1990. During both years a widespread St. Louis encephalitis epidemic was recorded throughout the southern half of the state (Day 2001). The environmental conditions during both epidemic years were remarkably similar. For example, the water table depth profiles for the two epidemic years are shown in Figure 2. Water table depth is the depth of the water column below the surface of the ground and can serve as a proxy for estimating the abundance of *Cx. nigripalpus* oviposition sites (Day and Shaman 2008, Shaman and Day 2005). In Figure 2, the annual ground water profiles for the 1977 St. Louis encephalitis epidemic (top line), 1990 St. Louis encephalitis epidemic (bottom line), and mean (middle line) daily water table depths are shown. Water table depths are deepest during the dry season (January through May, Weeks 1-22). For the model shown in Figure 2 the Initial Dry Down (IDD) begins at Week 14 (the first week in April) and extends through the end of Week 21 (the end of May). Beginning approximately Week 22 (the end of May) the first wetting events of the season are reported (IWET = initial wetting). During the two epidemic years the IWET sequence was followed by a secondary dry down (SDD) (July and August, weeks 27-34) that was remarkably synchronous during both years. The

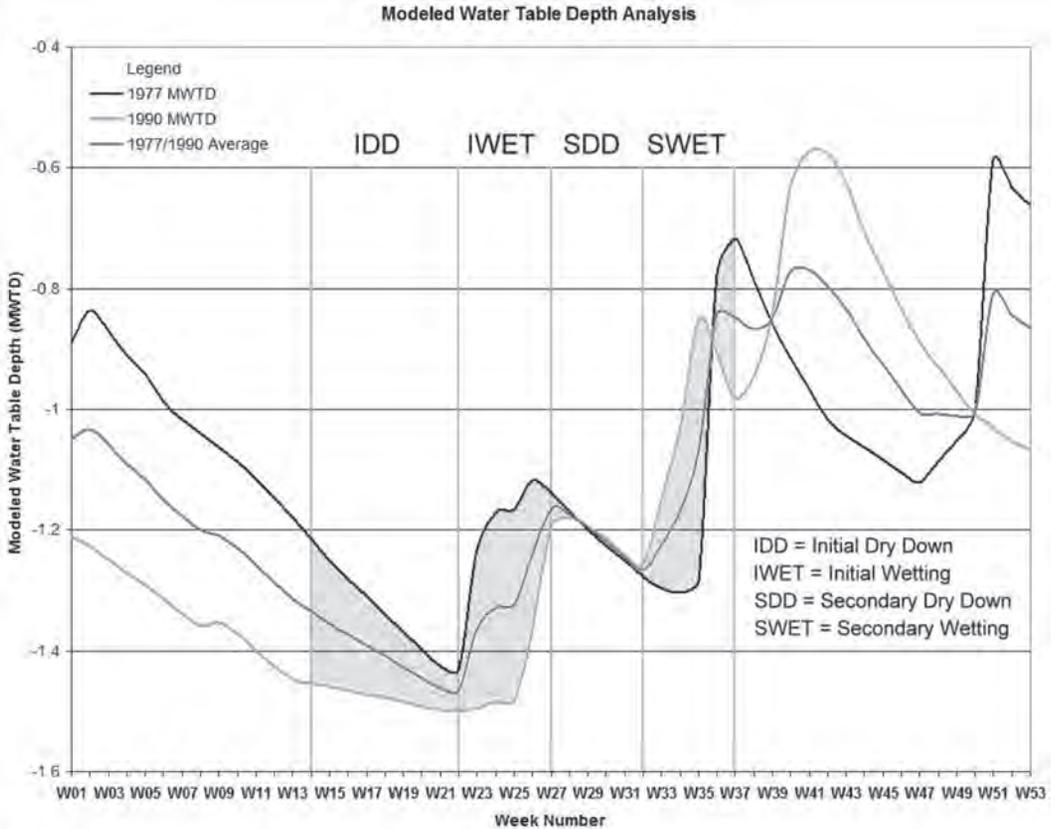


Figure 2. Modeled water table depth analysis for Indian River County during the 1977 and 1990 St. Louis encephalitis virus epidemics.

SDD was followed by a secondary wetting event (SWET) that coincided with the start of the south Florida rainy season (late August through early November, Weeks 35-44).

VI. ARBOVIRAL SURVEILLANCE AND PREDICTION

A thorough understanding of the factors influencing arbovirus cycles provides a means for predicting epidemics (Day 2001, Shaman et al. 2004, Day and Shaman 2011). By understanding the spatial and temporal distribution of the virus, the reproduction and age structure of vector and amplification host populations, and how environmental factors influence the cycling of the biotic components of mosquito-borne arboviral transmission cycles, it is possible to predict where and when arboviral epidemics will occur (Shaman et al. 2010).

In order to fully realize the ability to predict epidemics it is necessary to track components of all three biotic cycles in real time. It is important to have an understanding of spatial and temporal viral distributions. Sentinel surveillance is one of the best ways to track virus transmission through space and time (Day and Lewis 1992). Wild and domestic sentinels can be used to track viruses. Unfortunately, in many situations humans and horses serve as WNV and EEEV sentinels when the real goal is to predict these cases in advance. When this is the case, it is nearly impossible to use vector control and public health to get in front of a WN epidemic. By the time the first human cases are identified and confirmed, large numbers of humans are already infected.

Vector populations need to be tracked for abundance, spatial distribution, temporal distribution, reproductive history, population age structures, and (in some cases) mosquito

infection levels in order to assess real time amplification efficiency and transmission risk. Moreover, amplification host populations, their abundance, age structure, immune status, and reproductive success have to be tracked. In Florida, it is possible to conduct wild bird census surveys to track abundance, reproductive success, and age structures of known avian amplification host populations (Day and Stark 1999).

Finally, it is possible to track environmental parameters such as winter low temperatures and freezes, rainfall, water table depth, and ground surface dryness in real time to determine how these environmental factors may influence the reproduction and cycling of vector and amplification host populations. Together, these factors must be monitored and compared with well-established local baseline data to predict where the risk of arboviral amplification and transmission is high and where epidemic intervention in the form of vector control and/or a public health response is warranted (Day and Shaman 2011).

VII. CONCLUSIONS

1. All mosquito-borne arboviral transmission cycles consist of at least three biotic components: the arbovirus, one or more mosquito vector species, and one or more susceptible vertebrate host species.

2. Some arboviral transmission cycles (such as EEEV) involve secondary bridge vectors and avian amplification bridge hosts located in secondary amplification foci.

3. All of the Florida arbovirus transmission cycles are heavily influenced by abiotic environmental conditions, especially, but not limited to, rainfall and drought cycles, high summer temperatures, and winter subtropical freezes.

4. A thorough understanding of the biotic and abiotic factors associated with arboviral transmission cycles in Florida allows the prediction of where and when amplification may occur, the magnitude of that amplification, and whether the amplification has the potential to generate infectious mosquitoes in

numbers that will result in human epidemics and/or equine epizootics.

5. The occurrence of arboviral epidemics and epizootics in Florida are predictable. Well-designed surveillance strategies that generate long term local, or at the very least regional, baseline data need to be implemented and maintained with the understanding that epidemics and epizootics are rare events and that most of the surveillance data will be generated, analyzed, and catalogued during years of low arboviral activity.

VIII. ACKNOWLEDGMENTS

We thank James Newman for his work on Figure 1 and anonymous reviewers for their comments on early drafts of this manuscript. This work was supported by a long series of research contracts from the Florida Department of Agriculture and Consumer Services.

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MOSQUITO SPECIES COMPOSITION AND SEASONAL ABUNDANCE IN A NATIONAL ESTUARINE RESEARCH RESERVE IN NORTHEASTERN FLORIDA

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ABSTRACT. In October 2009, a mosquito surveillance program was established at the Guana Tolomato Matanzas National Estuarine Research Reserve (GTM NERR) in St. Johns County (northeast Florida). Mosquitoes were sampled monthly using CO₂-baited CDC traps from October 2009 through August 2012. During the 46-month study period, samples were collected for 25 months where a total of 11,460 mosquitoes, representing 23 species in 8 genera, were trapped. The three most abundant species were *Aedes taeniorhynchus* (31%), *Anopheles crucians* (20%) and *Culex nigripalpus* (15%). Of these three species, *An. crucians* was present in all but one month sampled (May 2012). Late summer through fall represented the period of greatest abundance for mosquitoes in GTM NERR. Further research in the reserve will allow for an even better understanding of northeast Florida mosquito communities' seasonal variation.

Key Words: estuarine, mosquitoes, trapping, surveillance

I. INTRODUCTION

Mosquito population surveillance is considered to be one of the most important activities performed by vector control agencies (Kline 2006). In October 2009, a mosquito surveillance program was established at the Guana Tolomato Matanzas National Estuarine Research Reserve (GTM NERR) in St.

Johns County (northeast Florida). Due to the popularity of the reserve for visitors and its large tracts of undeveloped land, public use is a primary focus. Unfortunately, large mosquito populations in the area can prohibit comfortable use of the reserve's facilities and trail systems. This has made routine surveillance of adult mosquitoes a necessity. Subsequently, data collected during these surveillance periods is shared with the Anastasia Mosquito Control District (AMCD) to supplement their larviciding efforts within the reserve. Due to strict regulations within the reserve boundaries, only the use of *Bacillus thuringiensis israelensis* (*Bti*), as a larvicide, is permitted. Therefore, surveillance via CDC trapping and physical inspection via airboat of the reserve's large impoundment (referred to locally as "Guana Lake") are mandatory functions conducted by GTM NERR and AMCD staff to prevent outbreaks of nuisance and vector mosquito species. Treating early instar larvae at the beginning of the peak mosquito season benefits control efforts by reducing the number of females that are capable of completing a gonotrophic cycle, however, the window of opportunity to successfully reduce mosquito larvae with *Bti* is very short.

One of the most common and highly targeted species is *Aedes taeniorhynchus* (Wiedemann), however, numerous *Culex* species along with *Ae. sollicitans* (Walker) can be problematic as well. With a fairly diverse number of larval production habitats available in the Reserve, numerous mosquito

populations may be present at any given time of the year. The goal of this study was to examine the seasonal species composition and abundance of mosquitoes in the coastal habitats of GTM NERR. Information on seasonal trends in mosquito frequency within the trapping area will allow for a broader understanding of the mosquito community within the Reserve, and potentially provide insight on mosquitoes in other coastal areas in northeast Florida.

II. MATERIALS AND METHODS

Adult female mosquitoes were collected in CO₂-baited CDC light suction traps from October 2009 through August 2012. Contents were collected monthly for only 25 months due to various reasons including trap failure, sampling scheduling conflicts, or inclement weather. For each sampling event, five CDC traps were placed on the GTM NERR peninsula, surrounding a freshwater marsh bordering Guana Lake. Trap locations were as follows (UTMs): Trap 1 (467641.9149, 3321260.67755), Trap 2 (467562.7724, 3321153.5674), Trap 3 (467612.7572, 3321013.134), Trap 4 (467430.6699, 3321231.5197), Trap 5 (467508.3248, 3321633.4803). Traps were deployed for approximately 16-18 hours then collected and contents taken to the GTM NERR laboratory, frozen for 30 minutes and identified using the identification key of Breeland and Loyless (1989). All mosquito species and additional trap contents were cataloged in an Excel spreadsheet and stored in a GTM NERR publicly shared folder. Other dipterans in trap collections were identified to family. For the purpose of this report only mosquito species were mentioned.

III. RESULTS

A total of 11,460 female mosquitoes, representing 23 species in 8 genera, were trapped in our study (Table 1). For each sampling period at least one species of mosquito was present in collections. Of the 23 species observed, the three most abundant species

were: *Ae. taeniorhynchus* (31%; n = 3,458), *Anopheles crucians* (Wiedemann) (20%; n = 2,228) and *Culex nigripalpus* (Theobald) (15%; n = 1,682). *Anopheles crucians* was the most frequently collected species and was present in all monthly collections except May 2012. The next most frequently trapped mosquito was *Cx. erraticus* (Dyar and Knab), trapped in 18 of 25 months. *Aedes taeniorhynchus* was collected 13 of 25 months, making it the third most frequently trapped species. The lowest number of species (one) in a collection period occurred on May 2012, while the greatest number (ten) occurred in the August 2012 traps. The months exhibiting the greatest diversity were late summer and fall months. October of each year (2009, 2010, 2011) exhibited the most diversity (excluding 2012). The greatest number of mosquitoes trapped (2,004 individuals) occurred in the July 2010 traps where *Ae. taeniorhynchus* accounted for >98% of the collection. The next two abundant months for mosquito collection were November and December of 2011, with 1,868 and 1,622 individuals trapped, respectively. Total abundance was greatest for the species mentioned above and occurred from July-December throughout the study period (Table 1).

IV. DISCUSSION

Within GTM NERR habitats, the most abundant species collected (regarding individuals caught) was not necessarily the most frequently occurring species within the CDC traps. This suggests that some species are moderately to highly seasonal and that ambient conditions, as well as other determining factors, may influence the presence or absence of any particular species at any single time in the GTM NERR. Environmental conditions certainly affect the presence of species. It could be possible that drought-like conditions during parts of 2011 and 2012 had some influence on the intensity of production for certain species as well as creating an overall shift in frequency of occurrence in traps. Such a scenario may explain why the number of species detected across each month remained relatively similar, while the

Table 1. Mean monthly abundance of mosquito species collected at Guana Tolomato Matanzas National Estuarine Research Reserve from CO₂-baited CDC suction traps from October 2009 through August 2012.

Species	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Monthly Average
<i>Aedes taeniorhynchus</i>	0	0	0	0	31.5	2.5	984.0	24.0	494.0	292.3	0.7	0.5	152.5
<i>Anopheles crucians</i>	61.5	65.4	93.0	34.0	36.0	7.5	112.5	162.5	59.0	83.3	98.7	244.5	88.2
<i>Culex nigripalpus</i>	0	0	0	0	0	0	8.0	92.0	0	142.0	352.0	0	49.5
<i>Cx. salinarius</i>	0	12.9	15.0	0	0	0	0	0	0	0	107.7	507.0	53.6
<i>Cx. erraticus</i>	14.0	9.0	28.0	18.0	1.5	1.5	3.5	13.0	8.0	217.7	32.0	164.5	42.6
<i>Cx. restuans</i>	26.5	30.2	26.5	0	0	0	0	0	8.0	12.0	82.0	181.5	30.6
<i>Cx. quinque.</i>	0	0	0	0	0	0	0	0	0	0	47.3	0	3.9
<i>Psorophora columbiana</i>	0	0	0	0	52.0	0	0.5	4.0	0	4.3	0	0	5.1
<i>Uranotaenia lowii</i>	0	0	0	0	0	0	0	0	0	24.7	0	1.0	2.1
<i>Ur. sapphirina</i>	0	0.4	0	0	0	0	15.0	1.0	0	1.7	0	16.0	2.8
<i>Ae. infirmatus</i>	0	0	0	0	0	0	0.5	18.0	0	8.7	0	0	2.3
<i>Ae. sollicitans</i>	0	0	1.0	0	2.5	1.5	13.5	2.0	0	2.3	0.3	0	1.9
<i>Culiseta. inornata</i>	2.5	4.4	0	0	0	0.5	0.5	0	0	0	0	0	0.7
<i>Ae. vexans</i>	0	0	4.0	0	0	1.5	0	0	0	0.3	0	0	0.5
<i>Ae. Mitchellae</i>	0	0	0	0	0	0	0	0	9.0	0.3	0	0	0.8
<i>Coquillettidia perturbans</i>	0	0	0	0	0	0	0.5	3.5	0	0	0	0	0.3
<i>An. quadrimaculatus</i>	0.5	0.4	0	0	0	0	0	0	0	0.7	0	0	0.1
<i>An. punctipennis</i>	1.0	0	0	0	0	0	0	0	0	0.3	0	0	0.1
<i>Cs. melanura</i>	0	0	0	0	0	0	0	1.0	0	0.3	0	0	0.1
<i>Ps. ciliata</i>	0	0	0	0	0	0.5	0	0.5	1.0	0	0	0	0.2
<i>Ae. canadensis</i>	0	0	0.5	1.0	0	0	0	0	0	0	0	0	0.1
<i>Ps. howardi</i>	0	0	0	0	0	0	0	1.0	0	0	0	0	0.1
<i>Cx. territans</i>	0	0	0	0	0	0	0	0	0	0	0.3	0	0.1
Mean totals/month	106.0	122.9	168.0	53.0	123.5	15.5	1138.5	322.5	579.0	791.0	721.0	1115.0	438.0
Species richness ¹	4.0	3.6	4.5	3.0	3.0	5.0	6.5	7.5	6.0	8.0	4.3	4.5	5.0

¹Species richness is the mean number of species detected each month.

number of individuals collected per month displayed no distinct pattern. The only trend demonstrated regarding mosquito species seasonality was during the month of October within the years of 2009, 2010 and 2011. October collections exhibited the greatest species diversity, however, it should be mentioned that at the time of this report the October 2012 sampling had not yet occurred. If August 2012 is any indication of upcoming species levels ($n = 10$), October 2012 should hold similar results, thus suggesting this time of the year as the most diverse for mosquitoes. Overall, few seasonal patterns were discerned for mosquito species composition or abundance. In general, however, late summer through fall represented the period of greatest abundance for mosquitoes in GTM NERR.

The most frequently occurring species was *An. crucians*, which was detected in all but one sampled month. There were also steady populations of *Culex*; detected in all but one month. Species from the *Aedes* genus including *Ae. taenioryhynchus* were also collected frequently, but were not nearly as abundant as *Anopheles* or *Culex*. Intensified sampling and more diverse bait assemblies could potentially increase trap collection efficiency and provide a more detailed description of the dynamics associated with mosquito abundance and diversity in the GTM NERR. Also, an alternate method of sampling to verify the efficiency of CDC trap locations may also allow for more detailed account of mosquito populations in the area. Precipitation and tidal events within the re-

gion coupled with the management of the Guana Lake by the Florida Fish and Wildlife Conservation Commission will influence the future of this data collection. Marsh management will also determine if more intensified sampling is necessary. At the time of the present study, mosquito control efforts were focused only on the impoundment, located north of the water control structure on Guana River road. Standing water and stagnant pond-like areas located within the peninsula of the reserve are untreated and can become production hotspots for freshwater mosquito species if precipitation occurs during warmer months of the year. Continued sampling in these and other nearby habitats will provide detailed information on seasonal and inter-annual species composition and abundance patterns of mosquito populations in the GTM NERR.

V. ACKNOWLEDGEMENTS

Special thanks to Mike Smith for technical help. Also, many thanks to Wendy Eash-Loucks, Jaime Pawelek, and AMCD and GTM NERR employees for additional help and consultation.

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SURVIVAL OF *Aedes aegypti*, *Aedes albopictus*, AND *Culex quinquefasciatus* ON NATIVE PLANTS UNDER LABORATORY CONDITIONS

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ABSTRACT. Survival on native Florida plants by adult *Aedes aegypti*, *Ae. albopictus*, and *Culex quinquefasciatus* was observed in the laboratory. The first survival experiment evaluated native plants that were non-flowering during the study period: redbay (*Persea borbonia*), cherry laurel (*Prunus caroliniana*) and American beautyberry (*Callicarpa americana*). Mosquito survival was significantly influenced by plant species. *Aedes aegypti* demonstrated a significant increase in survival at 72 hours on redbay, compared with the other two mosquito species. All three mosquito species survived well on cherry laurel with greater than 87% survival at 72 hours. A second experiment was conducted using only flowering plants: tall elephantsfoot (*Elephantopus elatus*), blanketflower (*Gaillardia pulchella*), spotted beebalm (*Monarda punctata*), and false indigo bush (*Amorpha fruticosa*). Significant differences of survival by species and by flowering plant were found. *Aedes albopictus* survived significantly better than the other two species on tall elephantsfoot. *Culex quinquefasciatus* survival was significantly lower on all four flowering plants compared with the other mosquito species. Overall, our findings highlight the complexity and importance of mosquito-plant interaction further supporting the need to understand mosquito sugar feeding behavior and how this obligatory behavior can be used for control.

Key Words: *Aedes aegypti*, *Aedes albopictus*, *Culex quinquefasciatus*, survival, plant feeding

I. INTRODUCTION

Mosquito control practices have historically consisted of targeting adults, when in

flight, by exposing them to an area-wide space spray of adulticides. Aerosolized droplets of the insecticide application must contact and subsequently be absorbed by the mosquito cuticle in order for toxicity to occur in the individual. More recently, application of boric acid sugar baits (Xue et al. 2006) and residual insecticides to non-flowering vegetation for control of adult mosquitoes have gained considerable attention (Qualls et al. 2012). This approach is based on the necessary resting behavior of these pests. Female and male mosquitoes obtain energy from carbohydrate sugars by feeding on floral nectar, nectaries on plant leaves and stems, and from homopteran honeydew (Yuval 1992, Foster 1995). Furthermore, because sugar-feeding behavior commonly occurs before and after a bloodmeal it is possible that a high percentage of individuals (nuisance and competent vectors) could be removed from the population, before their initial or next blood meal, by targeting this behavior (Foster 1995). Müller and Schlein (2008), Müller et al. (2008), and Müller et al. (2010) have developed a promising control method that exploits the dietary staples used to sustain the daily activities of mosquitoes through the use of attractive toxic sugar baits (ATSB). However, we need to further understand sugar feeding behavior and how native plants effect mosquito survival. This information could be used to determine if insecticide spot treatments to specific types of vegetation could benefit adult mosquito control. To address these issues we conducted laboratory tests on the survival of adult *Culex quinquefasciatus* Say, *Aedes albopictus* (Skuse) and *Aedes aegypti* (L.) on a variety of plants from their natural habitat in north-eastern Florida, specifically St. Johns County.

II. MATERIALS AND METHODS

Aedes aegypti, *Ae. albopictus*, and *Cx. quinquefasciatus* eggs were obtained from a laboratory colony maintained by the USDA-ARS, Center for Agricultural, Medical, and Veterinary Entomology, Gainesville, FL. Larvae of each species were reared in separate pans (50.8 × 63.5 cm) containing 600 larvae per 3,000 mL of reverse osmosis water. Each day until pupation, larvae were fed 1 g of ground up dog biscuits (Milk Bone™ Mini's Dog Treats, Del Monte Corporation). Mosquitoes were maintained under insectary conditions of 25°C, 85% relative humidity, and a photoperiod of 12:12 (Light: Dark) and provided a 10% sucrose solution.

All plant material was obtained from Anastasia State Park in St. Augustine, Florida. Non-flowering plants used in the first experiment were: redbay (*Persea borbonia* [L.]), cherry laurel (*Prunus caroliniana* [Aiton]) and American beautyberry (*Callicarpa americana* [L.]). All plants were identified by the St. John's County Agricultural Center, St. Augustine, FL. Plant cuttings were placed in a covered plastic cup (4 cm × 3.5 cm) in equal proportions, with a cotton ball saturated in water. Plant cuttings were then placed in cages made out of Rubbermaid® 1.9 liter screw-lid canisters. Approximately, 15 (three to five day old) females and 15 similarly aged males, sugar-starved for 48 hrs, were aspirated into each cage. The mosquitoes did not have access to water and were forced to feed on the plants for survival. Three cages with a 10% sucrose solution and no plant cuttings were set up in the same manner to serve as a positive control. Cages were then placed in the insectary and mosquito survival was recorded at 72 hours. Each plant species and mosquito species were replicated nine times.

A second experiment was conducted using only flowering plants and was conducted in the same manner as previously described. Flowers selected for this study were tall elephantsfoot (*Elephantopus elatus* Bertol.), blanketflower (*Gaillardia pulchella* [Foug.]), spotted beebalm (*Monarda punctata* L.) and false indigo bush (*Amorpha fruticosa* L.).

In both experiments mean percent survival of each mosquito species at 72 hours (corrected for control mortality using the formula by Abbott [1925]) on each plant species was subjected to analysis of variance. A Student-Newman Keuls (SNK) multiple range test was then performed on the arcsine transformed percent survival data (SAS Institute 2008). Mean survival by sex at 72 hours of each mosquito species by plant was analyzed using Student's t-test.

III. RESULTS

Mosquito survival was significantly influenced by plant species evaluated in experiment one ($F = 51.7$, $df = 1$, $P = 0.02$). Table 1 lists the mean percent survival times by species on each non-flowering plant species. *Aedes aegypti* demonstrated a significant increase in survival at 72 hours on redbay compared with the other two mosquito species ($F = 150$, $df = 1$, $P < 0.0001$). All three mosquito species survived well on cherry laurel with greater than 87% survival at 72 hours. *Culex quinquefasciatus* survival was significantly lower on the American beautyberry, compared with the other two mosquito species ($F = 27.8$, $df = 1$, $P < 0.0001$). Differences were also observed in male and female survival times by species on each plant where of all three species survived significantly better than females on redbay (Table 2).

Table 1. Mean ± SE percent survival of adult (mixed sex) *Aedes aegypti*, *Ae. albopictus*, and *Culex quinquefasciatus* observed at 72 hours exposure to various non-flowering plants collected from northeastern Florida.

Species	Redbay	Cherry Laurel	American Beautyberry
<i>Aedes aegypti</i>	77.4 ± 5.2 a*	92.5 ± 6.5 b	91.2 ± 8.4 b
<i>Aedes albopictus</i>	33.1 ± 4.9 a	99.1 ± 5.3 b	95.8 ± 6.2 b
<i>Culex quinquefasciatus</i>	22.5 ± 3.3 a	87.7 ± 4.6 b	51.2 ± 3.5 c*

Rows followed by different letters represent statistically significant differences on survival and plant species ($P < 0.05$) using SNK. *indicates a statistically significant differences on survival by mosquito and plant species ($P < 0.05$) using SNK.

Table 2. Mean ± SE percent survival of *Aedes aegypti*, *Ae. albopictus*, and *Culex quinquefasciatus* (separated by sex) observed at 72 hours exposure to various non-flowering plants collected from northeastern Florida.

Mosquito Species Tested	Plant Common Name	Plant Family	Male	Female
<i>Aedes aegypti</i>	redbay	Lauraceae	88.8 ± 3.1	63.3 ± 2.0 *
	cherry laurel	Rosaceae	95.7 ± 3.5	87.4 ± 3.0
	American beautyberry	Verbenaceae	94.6 ± 3.5	88.4 ± 3.2
<i>Aedes albopictus</i>	redbay	Lauraceae	38.7 ± 2.0	27.1 ± 2.4 *
	cherry laurel	Rosaceae	98.8 ± 3.7	99.4 ± 3.8
	American beautyberry	Verbenaceae	96.8 ± 3.6	94.9 ± 3.5
<i>Culex quinquefasciatus</i>	redbay	Lauraceae	25.3 ± 3.5	19.1 ± 2.7*
	cherry laurel	Rosaceae	87.2 ± 3.1	88.4 ± 3.5
	American beautyberry	Verbenaceae	48.0 ± 1.8	53.1 ± 2.4

*indicates a statistically significant differences on survival on the individual plant evaluated between male and female ($P < 0.05$) using Student's t-test.

There were significant differences on survival by mosquito species and flowering plant from the second experiment ($F = 20.7$, $df = 1$, $P = 0.02$). Table 3 lists the mean survival times by species on each of four flowering plants evaluated. *Aedes albopictus* survived significantly better on all of the flowering plants except for the blanketflower. *Aedes albopictus* survived significantly better than the other two species on tall elephantfoot ($F = 35.6$, $df = 1$, $P < 0.0001$). *Culex quinquefasciatus* survival was significantly lower on all four flowering plants compared with the other mosquito species. Survival of male *Ae. albopictus* was significantly greater on spotted beebalm than females ($t = 6.1$, $df = 15$, $P = 0.004$). No significant differences were observed in male and female survival times by the other mosquito species within plant species (Table 4).

IV. DISCUSSION

New approaches for integrated vector management methods are urgently needed (Townson et al., 2005, World Health Organization, 2012) and the awareness that some

plants result in increased survival could be a potential way of targeting control treatments or controlling mosquito populations by landscape management. Differences in landscaping and land use changes may attract different mosquito species including disease vectors. In this case, regional presence of actual (and competent) mosquito vectors may need to be considered. Müller et al. (2011) hypothesized that popular ornamentals pull mosquitoes like *Ae. albopictus* closer to domestic and peridomestic settings for sugar meals. A combination of toxic sugar baits and landscaping with unattractive plants that equal a push-pull strategy could reduce the density of container inhabiting mosquitoes. Utilizing mosquito resting and sugar feeding behavior, including landscape management, has direct application in mosquito control and needs to be further explored.

V. ACKNOWLEDGEMENTS

This is a research report only. We thank Katie Cash for her time in conducting these experiments.

Table 3. Mean ± SE percent survival of adult (mixed sex) *Aedes aegypti*, *Ae. albopictus*, and *Culex quinquefasciatus* observed at 72 hours exposure to various -flowering plants collected from northeastern Florida.

Species	False Indigo	Elephantfoot	Spotted Beebalm	Blanketflower
<i>Aedes aegypti</i>	72.1 ± 2.2 a	45.4 ± 1.5 b	70.2 ± 2.0 a	49.4 ± 1.3 b
<i>Aedes albopictus</i>	90.3 ± 3.2 b	97.0 ± 1.3 b*	94.2 ± 2.0 b	54.5 ± 2.6 a
<i>Culex quinquefasciatus</i>	26.8 ± 2.8 a*	30.2 ± 4.0 a	24.9 ± 2.7 a*	27.2 ± 1.6 a*

Rows followed by different letters represent statistically significant differences on survival and plant species ($P < 0.05$) using SNK. *indicates a statistically significant differences on survival by mosquito and plant species ($P < 0.05$) using SNK.

Table 4. Mean \pm SE percent survival of *Aedes aegypti*, *Ae. albopictus*, and *Culex quinquefasciatus* (separated by sex) observed at 72 hours exposure to various flowering plants collected from northeastern Florida.

Mosquito Species Tested	Plant Common Name	Plant Family	Male	Female
<i>Aedes aegypti</i>	false indigo	Fabaceae	73.6 \pm 2.4	70.8 \pm 2.4
	elephantfoot	Asteraceae	43.2 \pm 1.8	47.1 \pm 1.2
	spotted beebalm	Lamiaceae	72.3 \pm 2.4	68.7 \pm 2.9
	blanketflower	Asteraceae	48.6 \pm 1.7	50.3 \pm 1.8
<i>Aedes albopictus</i>	false indigo	Fabaceae	87.6 \pm 3.1	92.5 \pm 3.3
	elephantfoot	Asteraceae	100	95.2 \pm 3.0
	spotted beebalm	Lamiaceae	100	86.7 \pm 3.0*
	blanketflower	Asteraceae	52.5 \pm 3.2	55.8 \pm 1.8
<i>Culex quinquefasciatus</i>	false indigo	Fabaceae	26.5 \pm 2.5	28.0 \pm 4.2
	elephantfoot	Asteraceae	29.0 \pm 2.3	30.5 \pm 5.5
	spotted beebalm	Lamiaceae	23.6 \pm 3.9	27.2 \pm 2.6
	blanketflower	Asteraceae	30.5 \pm 2.3	26.0 \pm 1.5

*indicates a statistically significant differences on survival on the individual plant evaluated between male and female ($P < 0.05$) using Student's t-test.

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FIELD EVALUATION OF OCTENOL, LUREX 3, BG-LURE, AND OCTENOL+LUREX 3 COMBINATION AS BAITES IN BG-SENTINEL TRAPS TO COLLECT *Aedes albopictus*

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ABSTRACT. Octenol, Lurex 3, BG-Lure, and octenol +Lurex 3 combination as attractive baits in BG-Sentinel traps (BGS) and unbaited BGS traps were evaluated for their effectiveness to collect female *Aedes albopictus* in northeastern Florida from June 2010 through October 2011. During the study, seven species of mosquitoes (a total of 2,821) were collected, of which 97% were *Ae. albopictus* with 85% of that collection females. Octenol-baited BGS traps collected similar numbers of mosquitoes (46%), compared with traps baited with Lurex 3 (54%). Traps baited with octenol+Lurex 3 caught more mosquitoes (38%) than traps baited with octenol (25%), Lurex 3 (9%), or unbaited traps (27%). More mosquitoes were caught with BGS traps baited with the BG-Lure than those baited with Lurex 3. Our results showed that BGS traps baited with any of the above attractants were considered effective for the population surveillance of *Ae. albopictus*.

Key Words: BG-Sentinel trap, *Aedes albopictus*, octenol, Lurex 3, BG-Lure, attractant

I. INTRODUCTION

Traps combined with substances that act as attractants are effective tools for surveillance and control of vector and pest adult mosquitoes (Kline 2006). For example, BG-Sentinel™ traps (BGS) in combination with the BG-Lure™ (a mixture of ammonia, caproic acid, and lactic acid) have been documented as an efficient sampling tool for the dengue and yellow fever vector mosquito, *Aedes aegypti* (L.) and the potential dengue fever and chikungunya vector mosquito,

Ae. albopictus (Skuse) (Maciel-de-Freitas et al. 2006, Ritchie et al. 2006, Williams et al. 2006, Meeraus et al. 2008; Farajollahi et al. 2009, Lacroix et al. 2009, Ball and Ritchie 2010). Octenol (1-octen-3-ol) (Takken and Kline 1989, Kline 1994) and Lurex 3™ (lactic acid) have been evaluated as mosquito attractants in Mosquito Magnet® traps; the latter product proved to be a better attractant for collecting *Ae. albopictus* (Hoel et al. 2007, Xue et al. 2010). The BG-Lure, octenol and CO₂ have been tested as attractants in BGS traps to collect more vector mosquitoes (Irish et al. 2008, Bhalala and Arias 2009). To date, there is no published information about the effectiveness of Lurex 3 or its combination with octenol in BGS traps for collecting adult *Ae. albopictus*.

II. MATERIALS AND METHODS

Lurex 3 (a solid matrix of 35.4% AI lactic acid) and Mosquito Magnet-octenol sachet (solid polyethylene matrix containing 55.15% AI octenol) were both purchased online from Woodstream Corporation through www.mosquitomagnet.com. BG-Lures (a proprietary mixture of lactic acid, ammonia, and caproic acid in polyethylene matrices), were purchased from BioGents AG, Regensburg, Germany. BG-Sentinel traps (BGS) were used exclusively in this study and were purchased from BioGents AG.

For this study, the black catch pipe of the BGS trap was modified by attaching a kill tube (22 cm D × 11.5 cm H) (home made) with a screened bottom for air flow. By introducing a kill tube and adding a piece (3 × 4 cm) of the pesticide dichlorvos (AI 18.6%) strip (purchased from WalMart) specimens were less likely to escape when removing the

collection pipe. Field trials were conducted from June through October 2010-2011. The study site consisted of a large used tire pile (16 m W × 133 m L X 4 m H) in a rural area of Elkton, Northeast Florida (29.48.00N, 81.26.29W). Approximately a few thousand used tires were located on the study site. The surrounding area consisted of agricultural land with shallow ditches and an animal shed which contained 20-30 dogs. Based on preliminary BGS trap data, *Ae. albopictus* represented 97% of the population generated by the tires.

In the first experiment, three traps were baited with Lurex 3 and another 3 traps were baited with octenol. In the second experiment 2 traps baited with octenol, 2 traps baited with Lurex 3, and 1 trap baited with octenol+Lurex 3 were used, while one unbaited trap served as a control. In the third experiment, three traps were baited with BG-Lures and another 3 traps baited with Lurex 3. The distance between each trap, in all trials, was 10 meters and traps were left in the field for 24h. Each experiment was repeated on three separate occasions and all traps were rotated using a Latin square study design. The species and number of mosquitoes collected in each trap were separated by sex, identified, and counted.

Data obtained from each experiment were analyzed separately. Mean numbers of female *Ae. albopictus* collected in the BGS traps, for each attractant, were analyzed by one-way ANOVA. (Differences were considered significant when $P < 0.05$) Due to low counts of other mosquito species, their data was not used in statistical analysis.

III. RESULTS AND DISCUSSION

A total of 2,821 mosquitoes, representing 7 species in four genera, was collected during the study. *Aedes albopictus* represented 97% of the collections (2,443 females and 298 males). The other 3% (80) of the collections were *Anopheles crucians* Wiedemann, *Ae. infimatus* Dyar and Knab, *Ae. mitchellae* Dyar and Knab, *Culex erraticus* Dyar and Knab, *Cx. nigripalpus* Theobald, and *Psorophora columbiae* Dyar and Knab.

Table 1 shows that BGS traps baited with Lurex 3, or octenol, collected 7-8 times more

Table 1. Mean number of adult *Aedes albopictus* caught by BG-Sentinel traps baited with either octenol, or Lurex 3 in Elkton, FL (June–October 2010-2011).

Sex	Octenol	Lurex 3
Male	22 a	20 a
Female	142 a	173 a
Total (% of collection)	164 (46%)	193 (54%)

Means followed by the same letters in a row are not significantly different ($P > 0.05$)

female *Ae. albopictus* than males. There were no significant difference between the number of males collected in traps baited with Lurex 3 compared with traps baited with octenol; this trend continued for females as well. Moreover, slightly more individuals of both sex were collected with Lurex 3 (54%) compared with octenol (46%). The BG-Lure (60%) attracted significantly more *Ae. albopictus* than were collected by Lurex 3 (40%) (Table 2). BGS traps baited with or without attractants collected significantly more females than males of *Ae. albopictus* (Table 3). Overall, no significant differences existed between attractants evaluated, but traps baited with the combination of Lurex 3 + octenol collected the greatest number (38%) of mosquitoes, while the trap baited with Lurex 3 only collected the least number (9%). Traps baited with octenol only collected similar numbers of mosquitoes (25%) as unbaited traps alone (27%).

Lurex 3 contains lactic acid and was designed to be an effective attractant for collection of *Ae. albopictus* using Mosquito Magnet traps (Hoel et al 2007, Xue et al. 2010). The BG-Lure contains a combination of substances found on human skin, including ammonia, lactic acid, and fatty acids that has been demonstrated as highly attractive to *Ae. albopictus* (Meeraus et al. 2008). Although

Table 2. Mean number of adult *Aedes albopictus* caught by BG-Sentinel traps baited with BG-Lure and Lurex 3 in Elkton, FL (September-October, 2011).

Sex	BG-Lure	Lurex 3
Male	9 a	4 a
Female	203 b	140 c
Total (% of collection)	212 (60%)	144 (40%)

Means followed by the same letters in a row are not significantly different ($P > 0.05$).

Table 3. Mean number of adult *Aedes albopictus* caught by BG-Sentinel traps baited with either octenol, Lurex 3, octenol+Lurex 3, or unbaited in Elkton, FL (June-October, 2010-2011).

Sex	Octenol	Lurex 3	Octenol+Lurex 3	Unbaited trap
Male	12 a	5 a	12 a	15 a
Female	40 b	14 b	66 b	40 b
Total (% of collection)	52 (25%)	19 (9%)	78 (38%)	55 (27%)

Means followed by the same letters in a row are not significantly different ($P > 0.05$).

our study did not show significant differences between the different attractants when using BGS traps, the combination of Lurex 3 and octenol collected the greatest number of mosquitoes.

IV. ACKNOWLEDGEMENTS

The authors thank W. Qualls for statistical analysis. This is a report on a research study only and does not confer endorsement of any products by Anastasia Mosquito Control District.

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EVALUATION OF A PORTABLE BACKYARD MOSQUITO MISTER AGAINST *CULEX PIPIENS* AND *AEDES ALBOPICTUS* IN ISRAEL

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ABSTRACT. The magnitude of mosquito reduction and coverage area of the Terminix ALLCLEAR Backyard Mosquito Mister, a portable backyard mosquito mister, was evaluated using ALLCLEAR Naturals (geraniol and citric acid) and ALLCLEAR Synergized Plus (permethrin+piperonyl butoxide) products against host-seeking *Culex pipiens* and *Aedes albopictus*. The application dispersal pattern of the mister unit for a multi-directional and single-directional wind scenario resulted in a coverage area that far exceeded the product manufacturer's listed coverage area. Overall biting pressure reduction at the periphery of 200 m² and 300 m² experimental plots was significantly reduced for both species with both products. In light of these results, the backyard portable mosquito mister can be considered an alternative to stationary systems applying the same products with multiple nozzles that may be aesthetically unappealing and/or economically unfeasible to the homeowner.

Key Words: *Culex pipiens*, *Aedes albopictus*, misting system, geraniol

I. INTRODUCTION

Globally, mosquitoes are a nuisance and public health concern because they are im-

portant vectors of the pathogens responsible for malaria, West Nile fever, and dengue (Eldridge & Edman 2004, Service 1993). Mosquito population control and personal protection methods are the best measures to protect against mosquito-borne infection due to a lack of effective medical treatments for some of these diseases (Curtis 1992, Gupta and Rutledge 1994). Consequently, numerous products that reduce the biting pressure and increase personal protection, such as repellents and traps, are currently on the market (Revay et al. 2012). One method that has a considerable amount of attention by manufacturers and pest control professionals is the misting system. These automatically timed systems provide an envelope of protection against host-seeking mosquitoes within a defined area, e.g. residential backyards (Cilek et al. 2008).

Misting systems provide an alternative to using topically applied repellents because one of the most commonly used products, DEET (N,N-diethyl-3-methylbenzamide), has been reportedly linked with neurotoxic and allergenic effects in humans (Osimitz & Murphy 1997, Qiu et al. 1998). Although many misting systems are programmed for automatic application of an adulticide they can be manually turned on by the user during peak mosquito activity, e.g.. dusk and dawn, or when the user is outdoors poten-

tially providing protection from host-seeking mosquitoes in the immediate area.

While the use of automated systems for protection against host-seeking mosquitoes seems like a viable alternative, there has been very little scientific data to support their use in residential areas. The majority of commercially available systems consist of a series of multiple spray nozzles connected through a continuous loop of tubing to a reservoir tank containing an insecticide. However, new portable devices are currently on the market that may potentially provide the same type of application without the need for installation of a stationary system which may be aesthetically unappealing and/or economically unfeasible to the homeowner. One such portable product is the Terminix® ALLCLEAR Backyard Mosquito Mister, a device that claims to provide control of mosquitoes for up to 190 m². Thus, the purpose of this study was to determine the magnitude of mosquito reduction (and associated coverage area) provided by this product when applying either ALLCLEAR Naturals (active ingredients, AI, geraniol and citric acid) or ALLCLEAR Synergized Plus (AI permethrin plus piperonyl butoxide) against host-seeking *Culex pipiens* L. and *Aedes albopictus* (Skuse).

II. MATERIALS AND METHODS

Tests were performed in the northern Mediterranean coastal plain of Israel in suburban Haifa. The study took place from mid-June to mid-July 2012 just after sunset from 20:00 to 22:00. Weather conditions consisted of clear skies with early evening temperatures ranging from 27° to 30°C. Air movement was verified by smoke cartridges at a distance of 100 m to the experimental set ups and carefully observed during the experiments (air speed 0-8 km/h and non-directional). No unfavorable weather conditions were observed during the trial periods.

The equipment tested was Terminix ALLCLEAR Backyard Mosquito Mister (Figure 1), with either ALLCLEAR Naturals (AI 6.8% geraniol, 0.7% citric acid) or ALL-

CLEAR Synergized Plus (AI 10% permethrin, 10% PBO). The test unit was charged and operated as suggested by the manufacturer. Products were stored less than one month in the laboratory at ambient room conditions before they were tested.

Quantification of the mist dispersal pattern covered by one application of the unit was evaluated in open parkland under single and multi-directional wind conditions. Clear Din A-4 plastic sheets (3M, PP2200, St. Paul, MN) were mounted on wooden poles, 0.5m above the ground, with individual sheets positioned on the far side of 3m × 3m plots in a 30m × 30m square with the unit in the center. To facilitate observation of the mist dispersion pattern, we added 1% blue food dye to pure water. After one 4 min standard spray application, sheets were recovered and examined with a magnifying glass (3x) for droplet coverage. Sheets with 10 or more droplets were regarded as positive. The procedure was repeated 6 times. Single direction wind conditions were assessed similarly but the experimental set up was slightly modified with the unit upwind at the periphery of a 42 m × 42 m square, allowing wind to carry the mist over the test area.

Evaluation of mosquito efficacy (i.e. biting pressure) was conducted within a residential backyard (450 m²) in suburban Haifa. This area was largely protected from strong air movement by buildings and vegetation. In accordance with US EPA test guidelines, the study site exhibited a minimum biting pressure of 1 bite/minute of at least two different mosquito genera (EPA 1999). Common nuisance mosquitoes at the experimental and nearby control sites were *Cx. pipiens* and *Ae. albopictus*. Treatment and control sites were separated by 800m. Each product was evaluated under multi directional wind conditions at the periphery of areas of 200, 300 and 400 m² plots with the unit in the center of the yard. ALLCLEAR Naturals concentrate evaluation commenced two hours after a single mist application, and for ALLCLEAR Synergized PLUS concentrate six hours after application. The difference in



Figure 1. Terminix® ALLCLEAR Backyard Mosquito Mister and spray plume.

time intervals for testing was based on the manufactures instructions for each product. Similar evaluations occurred in non-treated control plots with the unit dispersing a water mist without active ingredients.

Six volunteers, two females and four males, were enlisted for this study. The volunteers were fully informed of the nature, objective, and procedures of the test including any physical and mental health conse-

quences that were reasonably foreseeable as a result of exposure to the test products. Tests were conducted according to EPA guidelines (EPA 1999). During evaluation, participants were not informed which mister was delivering active material. Volunteers were seated in chairs, as motionless as possible, facing towards the mister with one arm extended at a 45-degree angle, resting on thighs, in front of them. One forearm was exposed while suitable clothing protected the rest of body. In each trial, all volunteers rotated two times through single test stations and the control; $n = 12$ for 200, 300 and 400 m over three consecutive days. Each evening, one of the distances was tested; landing rates were evaluated for five minutes which enabled the group of volunteers to finish one trial in a half hour. Mosquitoes that either attempted to land, probe, and/or bite a volunteer's forearm were collected by assistants using hand nets and later recorded on data sheets. Assistants were fully protected by clothes and a topical repellent (Deepwoods Off!, AI 23.8% DEET, SC Johnson, Racine, WI) while standing behind the volunteers at a distance of about 1 m. A garden lantern (50 Watt, 8 feet distance from each collection site) provided adequate lighting during testing to enable the volunteers and assistants to observe any mosquitoes attempting to land.

Mean biting reduction at each distance between the different products and con-

trols were compared using Student's t-test (GraphPad Software Inc., La Jolla, CA).

III. RESULTS AND DISCUSSION

Dispersal pattern of the Terminix ALLCLEAR Backyard Mosquito Mister during multi directional wind conditions resulted in an average mist coverage area of 540 ± 8.1 m². The single directional wind evaluation resulted in an average coverage area of 610 ± 13.3 m². Both wind evaluations resulted in a coverage area that far exceeded the product manufacturer's listed coverage area of 190 m².

Mosquito biting pressure at the control site was considered high during the entire testing period and ranged from 11.78–21.89/5 minutes for *Cx. pipiens* and 9.94–17.39/5 minutes for *Ae. albopictus*. We considered that the overall biting pressure reduction at the periphery of our 200 m² experimental plot to be significantly effective with ALLCLEAR Naturals ($t = 13.2$, $df = 1$, $P = 0.04$) and ALLCLEAR Synergized Plus ($t = 14$, $df = 1$, $P = 0.05$) compared with controls (Table 1). Moreover, the unit provided significantly greater reduction of biting pressure with either active ingredient at the periphery of 300 m² plots (ALLCLEAR Naturals $t = 12.2$, $df = 1$, $P = 0.05$; ALLCLEAR Synergized Plus $t = 11.3$, $df = 1$, $P = 0.03$) compared with controls. At the periphery of 400 m² plots the total biting pressure was still

Table 1. Mean percent reduction of *Culex pipiens* and *Aedes albopictus* biting pressure¹ after using two products² at different distances when applied by the Terminix ALLCLEAR Backyard Mosquito Mister.

Mosquito species	Distance (m)	% Reduction		Untreated Control (raw means only)
		ALLCLEAR Naturals	ALLCLEAR Synergized Plus	
<i>Culex pipiens</i>	200	91.4	91.6	21.8
	300	65.6	69.8	11.7
	400	52.4	55.7	13.6
<i>Aedes albopictus</i>	200	82.1	87.2	17.3
	300	58.6	62.6	9.9
	400	46	42.5	15.5
Combined species	200	87.2	91.7	39.2
	300	62.4	66.5	21.7
	400	49.1	48.7	29.2

¹Biting pressure evaluated at 5 min intervals.

²ALLCLEAR Naturals evaluated at 2h, ALLCLEAR Synergized Plus evaluated at 6h.

reduced by nearly 50%. Currently accepted manufacturer guidelines require candles, coils, vaporizing mats, or other such products to provide at least a 50% repellency rate to make a reliable claim that the product repels mosquitoes (Govere and Durrheim 2007). Therefore, the effective coverage of the Terminix ALLCLEAR Mosquito Mister, in our study, exceeded the manufacturer claims of approximately 200 m².

We support the use of the ALLCLEAR Naturals product containing geraniol in the mister. A previous study evaluating another formulation of geraniol in a Terminix® ALLCLEAR Mister Lantern, demonstrated 80% mosquito biting reduction up to 91m² comparable with our ALLCLEAR Naturals application (Revay et al. 2012). Geraniol, a plant-derived alcohol, is considered completely safe for use and appears on the US Food and Drug Administration “Generally Regarded as Safe” list and is classified by the US EPA as a minimum risk pesticide under section 25(b) of the Federal Insecticide, Fungicide, and Rodenticide Act (EPA 2012).

IV. ACKNOWLEDGEMENTS

This research study was conducted according to protocol number AMCD 10-13-2005 as approved by the Anastasia Mosquito Control Board of Commissioners for use of human subjects in this project. Volunteers gave informed consent prior to participation in the study. The authors and AMCD do not

endorse any of the products evaluated in this report for the control of mosquitoes.

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LABORATORY EVALUATION OF SEVEN COMMERCIAL BARRIER TREATMENT PRODUCTS AGAINST *Aedes albopictus*

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ABSTRACT. Seven commercially available barrier application products sprayed on plants against introduced adult *Aedes albopictus* in laboratory cages was evaluated. The products were: Cy-Kick CS, Masterline, Tempo SC Ultra, Demand CS, Suspend CS, Talstar P, and Mavrik Perimeter. Residual leaf toxicity of all products, with the exception of Mavrik Perimeter, resulted in complete control of *Ae. albopictus* through four weeks and >90% mortality for the remaining two weeks afterward. Residual leaf toxicity from leaves treated with Mavrik Perimeter resulted in complete control of this species for two weeks immediately post treatment and steadily decreased to about 35% for the remainder of the six week study. At six weeks post treatment, mortality of products against male and female mosquitoes on treated leaves was compared; no product-related significant differences occurred. The results of this study demonstrate that there are many viable barrier application products that can provide up to at least 6 weeks control of *Ae. albopictus* adults in the laboratory making this alternative method potentially appealing to mosquito control programs for their use in field situations.

Key Words: *Aedes albopictus*, insecticides, vegetation, barrier treatment

I. INTRODUCTION

Treating vegetative perimeters, or artificial substrates, with residual insecticides to provide protection from nuisance and vector mosquitoes has become an option for homeowners and public health control professionals (Allan et al. 2009). These applica-

tions create an insecticide barrier between the mosquito population and local environs of a community (Perich et al. 1993). Barrier applications also target the resting behavior of mosquitoes which have been shown to rest in greater abundance in areas with greater vegetative cover (Bidlingmayer 1971).

Aedes albopictus (Skuse) is a primary nuisance mosquito in the southeastern United States (Nasci 1995) and the greatest source of service requests from residential areas in St. Johns County, FL (Xue unpubl.). Current strategies for *Ae. albopictus* control mainly rely on source reduction by emptying water-holding containers that serve as larval developmental sites and by using larvicides in those sites where removal or emptying is not feasible. Although reduction of larvae in container habitats may be immediate by using the methods above, the residual adult population takes more effort and a longer time to be controlled (Zhou et al. 2009). A wide range of pesticides have been evaluated as barriers when applied as residual foliar applications (Allan et al. 2009) against a variety of adult mosquitoes. Species differences in degree of knockdown and mortality after exposure to several active ingredients used for barrier application have been reported (Cilek and Hallmon 2008). Therefore, in order to determine the best active ingredient to achieve control of resting adult *Ae. albopictus* in vegetation we evaluated, in laboratory cage trials, seven commercially available products labeled for application as barrier sprays.

II. MATERIALS AND METHODS

Aedes albopictus eggs from a laboratory colony were obtained from the USDA-ARS,

Center for Agricultural, Medical, and Veterinary Entomology, Gainesville, FL. Larvae were reared in pans (50.8 × 63.5 cm) with 600 larvae per 3,000 ml of deionized water. Each day until pupation, larvae were fed 1 g of ground up dog biscuits (Milk Bone™ Mini’s Dog Treats, Del Monte Corporation). The colony was maintained under insectary conditions of 25°C, 85% relative humidity, and a photoperiod of 12:12 (light: dark) and provided a 10% sugar solution.

Table 1 lists each of the seven products evaluated. All products were received from Univar USA, Inc. (Austin, TX). Products were applied at maximum label rates. Seven 9.1L potted southern wax myrtle, *Myrica cerifera* (L.), were treated (one plant per product) until run-off using a 700 ml spray bottle. Controls were treated in the same manner with water. Plants were placed into individual cages in a Precision Incubator (Model 1818, Winchester, VA) set to 25°C, 85% RH, with a photoperiod of 12:12 (light: dark). Each plant was watered at its base every 24 hours to maintain plant health. Twenty-four hours post product application, 25 male and 25 female five to seven-day old mosquitoes were aspirated into each cage. Mortality was assessed at 48 hours. This evaluation was replicated three times for each product. Mosquitoes had access to a water-soaked cotton ball. Once weekly testing commenced thereafter for six weeks. Evaluation of each product and control was repeated three times at each time interval.

Average treatment mortality (corrected for control mortality using Abbott’s formula [1925]) was analyzed using a two-way ANOVA with treatment and time as factors with mean separation using Tukey’s studentized range honestly significant difference test (HSD) (SAS Institute 2008). Mean mortality of males and females at 6-weeks post application were compared within each product using Student’s t-test (P < 0.05).

III. RESULTS AND DISCUSSION

There were significant differences in *Ae. albopictus* mortality by week and treatment throughout the 6 week evaluation period

Table 1. Product name, % active and inert ingredients, manufacturer, and application rate evaluated in the laboratory against adult *Aedes albopictus*.

Product	% Active Ingredient	% Inert Ingredients	Manufacturer	Application Rate
Cy-Kick CS St. Louis, MO	6% cyfluthrin 59 ml per 304 m ²	94.00%	BASF, Corporation,	
Masterline	7.9% bifenthrin	92.10%	Univar, Austin, TX	14 ml per 304 m ²
Tempo SC Ultra	11.8% beta-cyfluthrin	88.20%	Bayer,	
Research Triangle Park, NC	16 ml per 304 m ²		Syngenta,	
Demand CS	9.7% lambda-cyhalothrin	90.30%		
Greensboro, NC	24 ml per 304 m ²		Bayer,	
Suspend SC	4.75% deltamethrin	95.25%		
Research Triangle Park, NC	44 ml per 304 m ²		FMC, Corporation Philadelphia, PA	29.5 ml per 304 m ²
TalStar P	7.9% bifenthrin	92.10%	Wellmark International, Schaumburg, IL	15 ml per 304 m ²
Mavrik Perimeter	22.3% tau-fluvalinate	77.70%		

Table 2. Corrected mean ± SE percent mortality of *Aedes albopictus* (mixed sex) by week after application of the various barrier treatment products.

Product	Percent Mortality (%) ± SE					
	WK 1	WK 2	WK 3	WK 4	WK 5	WK 6
Cy-Kick CS	100	100	100	100	99.3 ± 5.4	96.6 ± 5.5
Masterline	100	100	100	100	96.6 ± 2.5	93.3 ± 5.7
Tempo SC Ultra	100	100	100	100	96.6 ± 3.5	90 ± 2.3
Demand CS	100	100	100	100	98 ± 5.2	90 ± 6.4
Suspend CS	100	100	100	100	94 ± 4.7	90 ± 2.6
TalStar P	100	100	100	100	94 ± 3.1	90 ± 4.2
Mavrik Perimeter	100	100	84.6 ± 10.4*	64.6 ± 9.2*	55.3 ± 12.5*	35.3 ± 11.8*

*Treatment means in each column significantly different ($P < 0.05$) for insecticide and time interval (Tukey's HSD test).

($F = 30.2$, $df = 1$, $P = 0.003$). Residual leaf toxicity of all products, with the exception of Mavrik Perimeter, resulted in complete control of *Ae. albopictus* through four weeks and >90% mortality for the remaining two weeks afterward (Table 2). Residual leaf toxicity from leaves treated with Mavrik Perimeter resulted in complete control of this species for two weeks immediately after treatment then steadily decreased to about 35% for the remainder of the study. At six weeks post treatment, mortality of each product against male and female mosquitoes on treated leaves was compared where no product-related significant differences occurred (Table 3).

Overall, our results are consistent with other reports on semi-field and field barrier applications for control of mosquitoes. Royal (2004) reported that vegetation treated with bifenthrin resulted in a 94% reduction of native mosquito population in a residential neighborhood in Queensland, Australia, for six weeks. Residues of Suspend SC (deltamethrin) on treated leaves provided >95% overall knockdown/mortality up to 12 weeks

posttreatment (Cilek and Hallmon 2006). Another study found knockdown/mortality to be >95% at week 12 post treatment for products containing beta-cyfluthrin and lambda-cyhalothrin for both *Ae. albopictus* and *Culex quinquefasciatus* Say (Cilek and Hallmon 2008). Trout et al. (2007) reported that leaves treated with bifenthrin and lambda-cyhalothrin effectively controlled *Ae. albopictus* for six weeks after treatment. Also a recent field study in China showed that vegetation treated with lambda-cyhalothrin controlled *Ae. albopictus* populations two months post treatment (Li et al. 2010).

Based on the nature of barrier applications, targeting the resting behavior of mosquitoes, may prove to be a cost effective method of reducing mosquito populations especially *Ae. albopictus* (Trout et al. 2007, Doyle 2007, Doyle et al. 2009). Indeed, a variety of commercial products are now available that provides up to at least 6 weeks control (including those observed in our laboratory) makes this method even more appealing to operational mosquito management programs.

Table 3. Corrected mean ± SE percent mortality of female and male *Aedes albopictus* at 6 weeks post barrier application of each of the products evaluated, n = 75.

Product	Female	Male
Cy-Kick CS	97.3 ± 1.5	94.6 ± 2.3
Masterline	94.6 ± 2.5	90.6 ± 3.4
Tempo SC Ultra	92.0 ± 3.6	88.0 ± 4.2
Demand CS	90.6 ± 1.3	89.3 ± 3.5
Suspend CS	89.3 ± 1.5	90.6 ± 2.1
TalStar P	86.6 ± 6.7	93.3 ± 2.6
Mavrik Perimeter	36.0 ± 13.9	22.6 ± 12.9

IV. ACKNOWLEDGEMENTS

This research was supported by UNIVAR USA, Inc. Anastasia Mosquito Control District does not endorse any of the products evaluated in this study.

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SUCCESSFUL APPLICATIONS OF BARRIER TREATMENTS USING BIFENTHRIN AGAINST MOSQUITOES IN ST. JOHNS COUNTY, FLORIDA, FROM 2006 TO 2009

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ABSTRACT. The efficacy of barrier applications of Talstar One® (7.9% AI bifenthrin) in four different types of habitats targeting nuisance and vector fresh-water and salt-marsh mosquitoes was evaluated over a four year period. Efficacy varied, but in all evaluations mosquito populations were effectively reduced up to three weeks post-treatment. Barrier applications were determined to be most efficacious where the targeted site was surrounded by perimeter vegetation, and the application carried out on that perimeter. Based on our field trials, barrier applications have been incorporated into Anastasia Mosquito Control District's control program and have been a cost savings to the program.

Key Words: Barrier treatment, bifenthrin, vegetation, *Anopheles crucians*, *Culex nigripalpus*, *Culex quinquefasciatus*, *Aedes albopictus*

I. INTRODUCTION

Barrier application of insecticides to vegetation is one method of adult mosquito control that targets the resting behavior of this life stage. Residual insecticides have been used to treat perimeters of vegetation or artificial substrates to provide protection from nuisance and vector mosquitoes (Cilek and Hallmon 2008, Trout et al. 2007, Doyle 2007, Cilek 2008, Britch et al. 2009, Doyle et al. 2009, Qualls et al. 2012). This method has become a common practice for adult mosquito management by public health and mosquito control professionals.

A wide range of pesticides has been evaluated as residual foliar applications (Allan et al. 2009) with many recent field studies evaluating the active ingredient (AI) bifenthrin

(Doyle 2007, Trout et al. 2007, Cilek 2008, Britch et al. 2009, Qualls et al. 2012). Bifenthrin, a pyrethroid that has a long residual efficacy on a variety of surfaces, is labeled for residential mosquito control (Ansari et al. 1986, Yadav et al. 1996). This insecticide has provided acceptable reduction of host-seeking mosquitoes for 4-6 wk after application to residential sites or parks (Cilek and Hallmon 2008, Trout et al. 2007, Doyle 2007, Cilek 2008, Doyle et al. 2009, Qualls et al. 2012). The aim of such applications is to create an insecticidal barrier between the mosquito population and the area within the community (Perich et al. 1993).

At Anastasia Mosquito Control District (AMCD), St. Johns County (SJC), Florida we have utilized bifenthrin as a barrier application to handle repeat customer service requests and to protect the public in large recreational areas within SJC. Here we report on the efficacy of bifenthrin barrier applications carried out in four different habitat types targeting nuisance and vector fresh-water and salt-marsh mosquito populations.

II. MATERIALS AND METHODS

The first barrier application evaluation was applied in June 2007 to a homeowner's backyard adjacent to the Intercostal Waterway located in Ponte Vedra, Florida (30.251674, -81.434289). The site was selected for this treatment based on the homeowner's complaints about large populations of nuisance mosquitoes and the high number of landing rate counts (LRC) at this location on tri-weekly bases from the years 2004-2006. The house was located directly northeast of a large salt marsh producing

Aedes taeniorhynchus (Wiedemann) and *Ae. sollicitans* (Walker). Surrounding this site were many vacation homes that go unattended for long periods each year. Containers around these homes were often found to hold water that contained larvae and adults of *Ae. albopictus* (Skuse). An application of 4 ounces of TalstarOne® (AI 7.9% bifenthrin, FMC, Philadelphia, PA.) using a 5.6 liter pump-up sprayer (ACE ½ gallon polypropylene sprayer) was used to treat 372 m² of the homeowner's surrounding vegetation. Application was made early in the morning with favorable environmental conditions (<25.5°C, 70-80% RH, no wind) by a licensed AMCD employee.

Biting activity just prior to application (0700 hr) was evaluated with six 1-minute LRCs; three in the front of the residence and three in the backyard. The LRCs were conducted by an individual exposing their right leg from the knee down and recording the number of mosquitoes landing. These six pre-treatment counts established the initial baseline used to determine efficacy. The same number of LRCs in the front and backyard of this house (conducted at same time as above) were performed 24 hr post-treatment and continued weekly for six weeks. Similar landing rates at an untreated residence, 300 meters from the treatment site, were used as a control. The following formula was used to determine percent efficacy:

$$\frac{(\text{mean pre-LRC} - \text{mean post-LRC}) / \text{mean pre-LRC} \times 100\%}{}$$

Leaf bioassays were conducted on randomly sampled bifenthrin-treated leaves taken directly after application and weekly for six weeks post-treatment. Southern wax myrtle (*Myrica cerifera*) was the predominant plant species used in testing. Bioassays consisted of aspirating ten female laboratory-reared, 5-7 day old *Ae. albopictus* into 9 oz. clear plastic cups (SOLO® Cup Company, Lake Forest, IL) each containing a leaf (leaf length approximately 1 to 5 cm). Cups were then secured with screen mesh and a rubber band. The treated leaf and mosquitoes remained in cups for 24 h, at which time mortality was recorded. Controls consisted

of a similar set of untreated leaves. All treatments and controls were replicated 3 times per time period. Mosquitoes had access to 10% sugar-water soaked cotton balls placed on the top of the screened cup.

The second barrier evaluation was conducted at a 19 hectare SJC recreational park in September 2008 (29.832697, -81.36158). The recreational site consisted of low-lying floodplain areas that become mosquito production sites after a heavy rain event. During 2008, Tropical Storm Fay inundated this site producing large populations of *Ae. atlanticus* (Dyar and Knab), *Ae. infirmatus* (Dyar and Knab), *Culex nigripalpus* Theobald, *Psorophora columbiae* (Dyar and Knab), and *Culiseta melanura* (Coquillett). The park is surrounded by a nature trail that provides thick vegetation cover conducive to an insecticide barrier application. The majority of the vegetation consisted of cherry laurel trees (*Prunus caroliniana*), southern wax myrtle bushes, and pockets of bald cypress trees (*Taxodium distichum*). The perimeter vegetation of the park, a total of 836 m², was treated with 9.2 ounces of TalstarOne using a pump-up sprayer mentioned earlier. Application was conducted early (0700 hrs) in the morning with favorable environmental conditions (<25.5°C, 70-80% RH, no wind) by a licensed AMCD employee. The control site was located 500 m from the treatment area. CDC light traps baited with dry ice were used to monitor changes in adult mosquito populations in the treated and control areas. Traps were operated from 1500 to 0700 hrs.

Plant bioassays were conducted as mentioned earlier from treatment and control areas except that ten laboratory reared *Cx. quinquefasciatus* Say 5-7 day old females were used. The predominant plant species tested were leaves from southern wax myrtle and cherry laurel (leaf length approximately 1 to 5 cm). Mortality was assessed 24 hr post-treatment and weekly thereafter for six weeks.

The third barrier evaluation was conducted in September 2008 at the St. Augustine Amphitheater, a SJC public theater that hosts concerts and weeknight movies. AMCD was concerned with consistent reports of >30

mosquitoes landing per min at that site. The perimeter vegetation of the amphitheater, a total of 557 m², and was entirely treated with 6.0 ounces of TalstarOne using a 5.6 liter pump-up sprayer as earlier mentioned. Application was made early in the morning (0700 to 0800 hrs) with favorable environmental conditions (<25.5°C, 70-80% RH, no wind) by a licensed AMCD employee. Four sites were selected for pre- and post-treatment LRCs to determine the application efficacy. One LRC was taken at each site immediately before application and 24 after treatment then weekly for three weeks by the same licensed AMCD applicator.

The fourth barrier application was made near an AMCD sentinel chicken site (29.880689, -81.282143) that had a number of seroconversions for eastern equine encephalitis virus. This area was surrounded by many concerned equestrian owners. The perimeter vegetation along a road that was adjacent to the chicken site and equestrian farms was selected for barrier application. The perimeter vegetation, a total of 1021 m², was treated with 11.0 ounces of TalstarOne using a flo jet pump calibrated at 1.5 LPM and a 40° flat fan nozzle mounted on an all terrain vehicle. Application was made early in the morning with favorable environmental conditions (<25.5°C, 70-80% RH, no wind) by a licensed AMCD applicator. Mosquito Magnet X-traps (MMX) baited with dry ice were deployed for 24 hr pre-treatment and 24 hr post-treatment to determine percent change in mosquito populations. In addition to MMX trap collections, CDC traps baited with dry ice, octenol, and light were also deployed 24 hr before and once a week after for six weeks following treatment to determine the efficacy of the application.

III. RESULTS AND DISCUSSION

At the residential barrier application site we observed a 75% reduction of landing rates from salt-marsh mosquito populations up to four weeks post-treatment. At six weeks post-treatment LRCs were still reduced by 35% compared with pre-treatment counts which were as high as 35/min. LRCs at the control site remained high and averaged ≥20/min throughout the six week evaluation period. Excised leaves from the treated area continued to provide nearly 90% mortality in the laboratory bioassays for up to three weeks post-treatment (Table 1).

Ten mosquito species from six genera were collected at the control and treated sites of the recreational park. Twenty-four hours before application, 381 mosquitoes were collected in light traps at the treatment site compared with 155 at the control site. Five weeks after application, 56 mosquitoes were collected in light traps at the treatment site compared with 589 in the control site. Control traps showed a continuous increase in mosquito populations over the evaluation period while 91% reduction of the mosquito population was achieved at five weeks (Figure 1). Leaf bioassays also demonstrated high mortality post-treatment (Table 1) with >83% mortality observed up through week six.

At the amphitheater barrier application site, mosquito populations were greatly reduced up to three weeks post-treatment (Table 1). Importantly, amphitheater staff commented that following the barrier application they noticed a substantial reduction in mosquito bites.

Four mosquito species were collected with the MMX trap at the AMCD sentinel chicken barrier application site pre- and

Table 1. Mean ± SE percent mortality (24 hr) of female *Aedes albopictus* and *Culex quinquefasciatus* on excised leaves treated with bifenthrin at a residential backyard and recreational park. At the amphitheater evaluation site, only mean landing rate reduction ± SE is reported for *Aedes taeniorhynchus* after bifenthrin application.

Evaluation Site	Mosquito Species	WK 1	WK 2	WK 3	WK 4	WK 5	WK 6
Residential Backyard	<i>Aedes albopictus</i>	100	93.7 ± 3.4	89.5 ± 3.2	73.7 ± 2.4	37.9 ± 1.9	9.0 ± 3.3
Recreational Park	<i>Culex quinquefasciatus</i>	100	100	83.3 ± 2.9	87.2 ± 3	83.3 ± 2.9	65 ± 2.5
Amphitheater	<i>Aedes taeniorhynchus</i>	67.5 ± 1	97.5 ± 1.2	100	*	*	*

*No data collected.

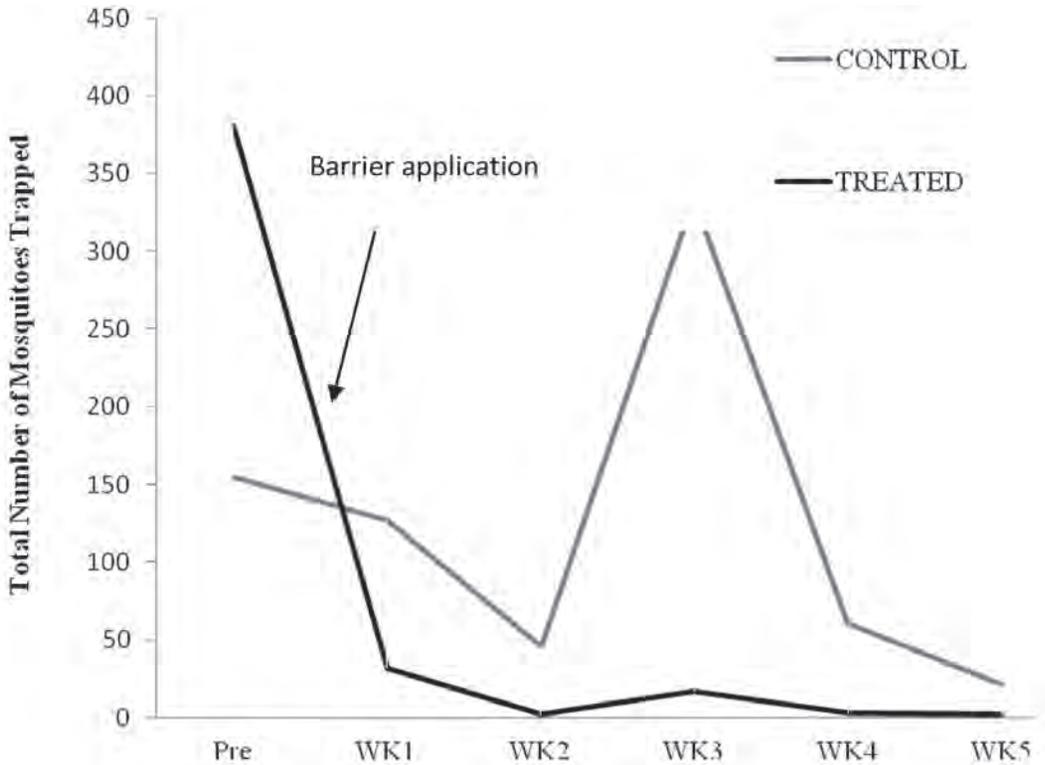


Figure 1. Total mosquitoes collected using octenol, light baited CDC traps at control and treated sites before and after barrier application at the recreational park.

post-treatment, these were: *Anopheles crucians* Wiedemann, *Cx. nigripalpus*, *Cx. quinquefasciatus*, and *Cs. melanura*. The overall reduction in mosquito populations was 90% up to six weeks post-treatment based on CDC light trap counts. *Anopheles crucians*, *Cx. nigripalpus*, and *Cx. quinquefasciatus* populations were reduced by greater than 40%. The important EEE vector, *Cs. melanura*, was reduced by 33% after the application. Prior to the barrier application there were 12 positive chickens for EEE; however, there were no additional positive seroconversions in AMCD sentinel chicken flocks at that site following treatment.

Generally, the barrier applications at all four study sites resulted in acceptable reduction in nuisance and vector mosquito populations for at least three weeks post-treatment. Although we used a variety of methods to determine efficacy of the barrier applications, the number of service requests,

number of mosquitoes landing per minute, and the number of mosquitoes collected in traps was reduced during these evaluations regardless of application site. The results of this study strongly support the implementation of barrier treatments into mosquito control operational programs.

IV. ACKNOWLEDGMENTS

The authors thank Tom Downey and Mike Mills for their valuable assistance in the field. This is a report on a research study and does not imply endorsement by Anastasia Mosquito Control District of St. Johns County, Florida for any products.

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FIELD EVALUATION OF THERMAL FOG APPLICATION OF SUMITHRIN PRODUCTS AGAINST ADULT MOSQUITOES IN NORTHEAST FLORIDA

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ABSTRACT. Thermal fog applications of Duet (a mixture of prallethrin, sumithrin, piperonyl butoxide [PBO]) caused significantly greater mortality (80-84%) in semi-field studies with caged *Aedes albopictus* and *Culex quinquefasciatus* compared with mortality (59-66%) caused by Anvil 10+10 (sumithrin+PBO). Mortality (76%) of caged *Ae. albopictus* caused by thermal fog application of Duet diluted by 50% with mineral oil was significantly greater than mortality (31%) caused by a Duet dilution of 75%. Natural populations of *Ae. taeniorhynchus* following thermal fog application of Anvil 10+10 ULV were reduced nearly 93% while natural populations of *Cx. nigripalpus* were reduced by 98.6% after undiluted Duet application in field trials.

Key Words: Sumithrin, thermal fog, *Aedes albopictus*, *Aedes taeniorhynchus*, *Culex nigripalpus*, *Culex quinquefasciatus*

I. INTRODUCTION

Sumithrin is a broad-spectrum pyrethroid insecticide that has been developed as the active ingredient in several commercial products, e.g., Anvil 10+10® ULV and Duet™. These products have been registered and are currently used for adult mosquito control by aerial and ground cold ultra-low volume (ULV) application including thermal fog in Florida (Meisch et al 2005, Qualls and Xue 2010, Xue et al. 2012). Since 1970, mosquito control programs have largely adopted the ULV technique for adult mosquito control (Mount 1998) effectively replacing, in most instances, traditional thermal fog application.

During diurnal periods, adult mosquitoes will often rest or hide in vegetation. Studies demonstrate that effectiveness and dispersal of insecticides by cold ULV are affected by the density of vegetation (Taylor & Schoof 1971, Curtis & Carlson 1990) and the environment (Britch et al. 2010). At the Anastasia Mosquito Control District (AMCD) the number of individuals requesting mosquito control by hand-held fog applications during the day has increased. Moreover, budget cuts have limited the number of nighttime ground ULV application. These factors compel us to re-evaluate the efficacy of hand held thermal fog application using current insecticide formulations applicable for this type of equipment. The purpose of this study was to determine if control of adult mosquitoes in forested/vegetated habitats could be achieved using Anvil 10+10 or Duet when applied by a hand held thermal fogger during diurnal mosquito resting periods (especially during normal daytime business hours at AMCD).

II. MATERIALS AND METHODS

Insecticides: Duet (A.I. 1% prallethrin, 5% sumithrin, 5% piperonyl butoxide [PBO]) and Anvil 10+10 ULV (A.I. 10% sumithrin, 10% PBO) were used in this study. Both insecticides were obtained from Clarke, Roselle, IL. In addition, two concentrations of Duet (2.5% and 0.5%) diluted in mineral oil were evaluated.

Semi-field test with caged mosquitoes: Laboratory-reared *Cx. quinquefasciatus* and *Ae. albopictus* were used for the semi-field test which were conducted at St. Johns Fairground (29 46' N, 81 27' W). Field-collected

Culex egg rafts and *Aedes* eggs from AMCD's operational area were reared to adults at the AMCD insectary. Five to 7-day old non-blood-fed females were utilized in these trials. Cylindrical cages fashioned from poly-chloride vinyl pipe (15 cm diam × 5 cm high) with both ends covered by white mesh screens (17 × 17 mm) were used in the bioassays. Ten female mosquitoes were transferred into each cage with a mouth aspirator. Three control and 9 treatment cages were used for each test. Treatments cages were set out 50 m apart at the top of 1 m poles in a 100 m × 100 m open field plot at the fairground. Control cages were exposed in the same location for 15 min prior to insecticide application, then immediately held in an enclosed vehicle.

A TS-35 model hand thermal fogger (American LongRay, San Francisco, CA) was used for application. Anvil or Duet was applied at an application rate of 34 ml/min by a trained AMCD employee 5 m from the first line of cages at a walking speed of 2 km/h while taking wind direction into account. After treatment, all cages were brought back to the laboratory, where mosquitoes were transferred to clean cages and provided with cotton balls of 10% sugar solution placed on top each cage. Mortality in treatments and controls were recorded 12 hours post-treatment. Three tests for each insecticide, or each formulation, were performed against caged mosquitoes.

During semi-field trials, air temperature ranged from 75-85° F during the 7 am to 11 am test period; the average relative humidity was 86%-98%, and wind conditions varied from 1-4 mph. These data were recorded at 15 min intervals throughout the test period using a portable weather station (MX 2000, Fischer Scientific).

Field tests with natural mosquito populations: Hand-held thermal fog application of Anvil 10+10 against natural populations of *Ae. taeniorhynchus* were conducted to vegetation along a forested area off Neck Road in Ponte Vedra and Cornerstone Park, St. Augustine, FL. Human landing rate counts were conducted on a human volunteer at 3 min pre- and post-application intervals. Four replications were taken at both intervals. In-

secticide was applied at the same flow rate and method as mentioned above for the semi-field trial. About 1,000 linear meters were treated at each site.

A third study evaluated the application of undiluted Duet against natural populations of *Cx. nigripalpus* in a forested area of Elkton, St. Johns County, at the same flow rate and method as mentioned above for the semi-field trial. Evaluation of efficacy used CDC traps baited with dry ice overnight at pre- and post-treatments each time. Application consisted of the sprayer walking a linear transect along the edge of the forested area for 1 km to allow the fog to drift into vegetation. Temperature, wind speed, direction, and relative humidity were recorded pre- and post-application using a hand-held digital device (Skywatch Xplorer, N Tech Holmen, WI).

For the semi-field test, mean percent mortality of each species was calculated without a correction factor because no mortality was observed in control cages. Mean percent reduction of natural mosquito populations in field trials was calculated based on the mosquito landing rate count (LRC) in pre-treatment divided into the LRC at post treatment. All data were analyzed by Student's t-test.

III. RESULTS AND DISCUSSION

The Duet application resulted in significantly greater ($t = 8.1714$, $df = 9$, $P=0.0001$) mortality for each mosquito species (80-84%) compared with Anvil (59-66%) but there was no significant difference within product between species (Table 1). Our results with thermally applied Anvil against

Table 1. Mean percent mortality at 24 hr post-treatment of caged *Culex quinquefasciatus* and *Aedes albopictus* following thermal fog applications of Anvil 10+10 and Duet at St. Johns Fairground, St. Johns County, FL.

Product	<i>Culex quinquefasciatus</i>	<i>Aedes albopictus</i>
Anvil 10+10	66 a	59 a
Duet	80 b	84 b

Paired t-tests were separately performed on data in columns and rows. Means followed by the same letter are not significantly different ($P > 0.05$).

Table 2. Mean pre- and post treatment landing rate counts and percent reduction of natural populations of *Aedes taeniorhynchus* following thermal fog application of Anvil 10+10 in Ponte Vedra (P.V.) and St. Augustine, FL.

Date and Site	Pre-treatment	Post-treatment	Landing Rate Reduction (%)
6/30/2011 P.V.	24.0	1.7	93.0
7/22/2011 P.V.	6.7	0.6	91.0
7/26/2011 St. Augustine	8.7	0.6	93.0
7/28/11 P.V.	27.0	1.7	94.0
Overall mean	16.6	1.2	92.8

caged *Cx. quinquefasciatus* and *Ae. albopictus* were similar to results from a previous report on ground ULV application of this insecticide against caged *Ae. sollicitans* (Walker) in semi-field trials (Lesser 2002).

Mortality of caged *Ae. albopictus* (76%) from the Duet formulation diluted by 50% with mineral oil was significantly greater ($t = 9.32$, $df = 3$, $P = 0.0026$) than the mortality (31%) caused by a dilution of 75%. The 50% application diluted from original stock concentrate still resulted in acceptable mortality. The per gallon cost for Duet stock is \$180; after 50% dilution the cost, including mineral oil, was reduced to about \$100.

In Anvil field trials, the overall *Ae. taeniorhynchus* LRC was reduced by nearly 93% (Table 2). Air temperature ranged from 80-90°F from 8am-11am, and wind varied from 0-5 mph during those evaluations.

In the field trial evaluating thermal fog application of Duet against natural mosquito populations in Elkton, 6,063 mosquitoes were collected by CDC traps baited with dry ice the night before treatment. *Culex nigripalpus* represented 85% of that collection, *Anopheles crucians* Wiedemann represented 14%, and all other species 1%. After treatment, 87 mosquitoes were collected by the traps, of which *Cx. nigripalpus* were 34%, *An. crucians* 57%, and other species 9%. Overall population reduction was 98.6%.

Historically, thermal or non thermal adulticide application has not been observed to provide significantly different mortality (Taylor & Schoof 1968, Mount et al. 1966). Standards for today's thermal fog equipment for vector control continue to evolve with several workers characterizing droplet size and dispersal patterns (e.g. Hoffmann et al. 2008). For what was old is now new. Therefore, it is appropriate that as new adulticide

formulations come into the marketplace for mosquito control that we consider adapting them in novel, as well as traditional ways for our operational programs.

IV. ACKNOWLEDGEMENTS

This research study was conducted according to protocol number AMCD 10-13-2005 as approved by the Anastasia Mosquito Control Board of Commissioners for use of human subjects in this project. Volunteers gave informed consent prior to participation in the study. The authors, or District, do not endorse any of the products evaluated in this report for the control of mosquitoes.

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FIELD EFFICACY OF GROUND ULV SPRAY OF AQUARESLIN® AGAINST CAGED *CULEX QUINQUEFASCIATUS* IN ST. AUGUSTINE, FLORIDA

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ABSTRACT. Efficacy of ultralow volume (ULV) ground application of AquaReslin (permethrin) against caged *Culex quinquefasciatus* was evaluated in 2 residential areas of St. Augustine, Florida. The night application did not result in significant mortality of caged mosquitoes on either side of the street, except for 38-40% mortality in cages placed at the turn point. Data analysis showed that insufficient wind speed was the primary reason for inadequate mortality. Best management practice for ground ULV application of mosquito adulticides was reviewed, revised accordingly, and adopted.

Key Words: ultra low volume, permethrin, *Culex quinquefasciatus*, pyrethroid insecticides

I. INTRODUCTION

Permethrin is a broad-spectrum insecticide that has been formulated as several commercial mosquito adulticides e.g. Aquasure®, AquaKontrol®, and AquaReslin®, (Qualls et al. 2011). AquaReslin is registered for adult mosquito control by ultra low volume (ULV) ground application in Florida (Xue et al. 2008). The technology of ground ULV applications of pesticides to control mosquitoes was pioneered in the early 1970's and widely adopted by most mosquito control programs in the USA (Mount 1998, Meisch et al. 2006, Xue et al. 2008). The dispersal of insecticides by cold ULV is limited by weather conditions, especially wind direc-

tion and speed. Indeed, product labels of mosquito adulticides formulated for ground ULV application have upper wind speed restriction thresholds. Normally, weather conditions are determined by daily operation supervisors, at the district level, 2-3 hours prior to spraying (application usually takes place well after normal work hours). However, change of local weather conditions in remote field sites can be difficult to predict. We conducted a routine field experiment to assess the effectiveness of AquaReslin ULV ground applications against caged adult *Culex quinquefasciatus* Say. During those applications suboptimal weather conditions occurred. As a result, we present our study results during those conditions and provide insight for refinements of Anastasia Mosquito Control District's (AMCD) adulticiding program.

II. MATERIALS AND METHODS

AquaReslin (AI 20% permethrin, 20% piperonyl butoxide) was used in all testing and obtained from Bayer Environmental Sciences, Montvale, NJ. The adulticide was applied by an AMCD certified spray technician using a ULV heavy-duty truck-mounted sprayer (Grizzly, Clarke, Chicago, IL) at an application rate of 0.9 g per acre. Droplet size of the adulticide with the ULV machine was previously calibrated to deliver a range of droplet size from 15 to 30 μ . Mosquito efficacy of the treatment was assessed against caged *Cx. quinquefasciatus*

using the methods of Amoo et al. (2012). Field-collected egg rafts from St. Johns County were reared to the adult stage at the AMCD insectary. Bioassay cages were made from polychloride vinyl pipe (15 cm diam × 5 cm high) with both ends covered with white screen mesh (17 × 17 mm) fastened by rubber bands. Fifteen, 7-day old non-bloodfed female mosquitoes were transferred into each cage using a mouth aspirator. Two locations were selected for the experiment. Cages were set-up on both sides of the street to avoid possible wind directional changes during application (i.e. cages from one side of the street would be exposed to the ULV spray regardless of wind direction). Each test consisted of 12 cages: three cages placed on one street side, another three cages placed on the opposite street side, and three cages placed at the corner of the street turn point. The distance between each cage on both side streets was 30 meters and 10 meters between each cage on the street turn point. The distance from truck spraying to the cage was about 15 meters. All cages were set up beside mail boxes in the open field. The remaining three cages served as untreated controls and were placed in the same neighborhood about 1 km from the treatment to avoid insecticidal drift from the application. Spray technicians were not informed of the experiment set-up. Vehicle speed was 15 km per hr. Wind direction and speed at the beginning of the application was recorded. After treatment, all cages were brought back to the laboratory, mosquitoes transferred to clean cages, and provided with cotton balls of 10% sugar solution placed on top each cage. Mortality in treatments and controls were recorded 12 hours post-treatment.

III. RESULTS AND DISCUSSION

Table 1 shows the mean percent mortality, at each location, from caged *Cx. quinquefasciatus* that resulted from the AquaReslin application. Data showed that 4.4% and 2.2% mortality occurred in cages placed on one side of the street, no mosquito mortality on

Table 1. Twelve hour post-treatment mean percent mortality of caged *Culex quinquefasciatus* (15 female mosquitoes/cage × 3 cages) after exposure to ULV applied AquaReslin at St. Augustine, FL.

Treatment location	Crescent Beach	Vilano Beach
One street side	4.4	2.2
Opposite street	0	0
Street turn point	40.0	38.0
Control sites	0	2.2

opposite street, and 40% and 38% mosquito mortality on the street turn point in the first and second experiment, respectively. The application did not result in an acceptable level of mortality at either test site. Moreover, residents in the area still complained about mosquitoes. In fact, the human landing rate count was still as high as before the application (i.e. 3-5 per minute). Spray parameters such as temperature, relative humidity, wind direction and speed were examined post-treatment. We found the wind speed (0-2 mph) was unsuitable at the time and location of application that lead to the ineffective result. The 38-40% mosquito mortality that occurred in the cages at the turn points may have resulted from casual wind/swirl possibly generated by the spray truck and additional vehicle traffic in the immediate area.

Lack of acceptable results from the above study generated a critical review of the District's best management practice for ground ULV application. Our adulticide program was rewritten to enhance monitoring of possible meteorological impacts on ground ULV application efficacy (Brown & Cope, 2004). As a result we now provide digital weather monitors to spray personnel to monitor wind speed and direction in the targeted application area. We also pay spray personnel overtime, collect post-treatment efficacy data, calibrate equipment every 50 hrs, monitor pesticide concentration in the truck-mounted tanks (Xue et al 2008), monitor for insecticide resistance, and promote barrier treatment of vegetation (Amoo et al 2008). Based on these changes and improvements, the effectiveness of our ground ULV treatments has been more efficient.

IV. ACKNOWLEDGEMENT

Anastasia Mosquito Control District does not endorse any of the products evaluated in this study.

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EFFICACY OF ALTOSID XR-G EXPOSED TO SUNLIGHT AND TEMPERATURE VARIATIONS AGAINST FOUR SPECIES OF MOSQUITO LARVAE

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ABSTRACT. This study evaluated the efficacy of Altosid® XR-G longevity when exposed to direct sunlight and to two temperature regimes against larvae of *Aedes aegypti*, *Ae. albopictus*, *Ae. taeniorhynchus*, and *Culex quinquefasciatus*. An average of 67.2% emergence inhibition was found for all species exposed to the product with 16 months exposure to sunlight and ambient temperature. Emergence inhibition was >75% for all three species to product that had been exposed to 15°C or 39°C for one month. Our study demonstrated that efficacy and persistence of Altosid XR-G can be affected by certain factors with potential implication for application effectiveness in areas that become inaccessible following a flooding event.

Key Words: methoprene, larvicide, temperature, sunlight, mosquitoes

I. INTRODUCTION

Altosid® XR-G (s-methoprene) is formulated as a granular sustained-release insect growth regulator. This product has effectively provided 30 days or longer control of floodwater and salt marsh mosquitoes ((Linthicum et al. 1989, Logan et al. 1990, Kramer and Beesley 1991, Qualls and Xue 2007). Additional benefit of the XR-G pellet is that it can be applied before a rain event as a pre-treatment/pre-flood application in areas that become inaccessible following rain or tidal events. Logan et al. (1990) demonstrated that when the product was applied 5 weeks prior to a rain event it resulted in 98% adult mosquito emergence inhibition (EI). Qualls and Xue (2007) showed 92% and 84% EI of salt marsh mosquitoes follow-

ing 50 day pre-treatment application at rates of 9.0 kg/ha and 4.5 kg/ha, respectively. However, longevity of the active ingredient after exposure to various environmental parameters such as sunlight and temperature is unknown. Therefore, the extended efficacious period of control (i.e. EI) is unclear for such scenarios. We report herein on our study that evaluated the EI longevity of Altosid XR-G when exposed to direct sunlight and varying temperature regimes against larval *Aedes aegypti* (L.), *Ae. albopictus* (Skuse), *Ae. taeniorhynchus* (Wiedemann), and *Culex quinquefasciatus* Say.

II. MATERIALS AND METHODS

Aedes aegypti and *Cx. quinquefasciatus* eggs were obtained from a laboratory colony at the USDA-ARS, Center for Agricultural, Medical, and Veterinary Entomology, Gainesville, FL. *Aedes albopictus* and *Ae. taeniorhynchus* larvae were collected from field sites around St. Johns County, Florida. Larvae of each species were reared separately in pans (50.8 × 63.5 cm) containing 600 larvae per 3,000 ml of distilled water fed 1 g of ground up dog biscuits (Milk Bone™ Mini's Dog Treats, Del Monte Corporation) daily until pupation. Adult mosquitoes were maintained under insectary conditions of 25°C, 85% RH, a photoperiod of 12:12 (L: D), and provided a 10% sugar solution.

A 0.19 kg sample of Altosid XR-G (AI 1.5% s-methoprene, Zoecon, Wellmark International, Schaumburg, IL) was placed in a white enamel rearing pan (50.8 × 63.5 cm) on the grounds of the Anastasia Mosquito Control District Base Station, St. Augustine, FL. The rearing pan was covered with clear Plexiglas to prevent wetting from rain and

remained outside to be exposed to natural sunlight and ambient temperatures for 16 months (October 2006-February 2008). At 1, 3, 10, 12, and 16 months, ten, third instar larvae of each of the above 4 mosquito species were individually placed into a 255 g cup containing 100 ml of deionized water and a quantity of solar-exposed XR-G pellets to achieve a 500 ppm concentration. Controls were handled similar but cups contained 100 ml of deionized water only. Testing was carried out in an incubator (Model 1818, Winchester, VA) at 12L:12D cycle and 28°C.

A separate aliquot of XR-G granules were exposed to 2 constant temperatures of 15° and 39°C, respectively, for 30 days in darkness in the incubator. Larval assays were conducted in the same manner as previously described. Experiments were repeated three times against third instar *Ae. albopictus*, *Ae. aegypti*, and *Cx. quinquefasciatus*.

In all tests, treatments and controls were replicated nine times per species at each time period. Each evaluation period was repeated three times. Percent emergence inhibition was calculated as follows:

$$\frac{([\text{total number individuals} - \text{emerged pupae}] \times 100)}{\text{total number individuals}}$$

Time period post-exposure within each species on mortality was calculated and analyzed using an ANOVA (SAS Institute 2008). Mean percent EI between treatment and controls were compared using the paired t-test.

III. RESULTS AND DISCUSSION

Table 1 shows the percent EI of four mosquito species treated with Altosid XR-G after

continuous solar exposure and ambient temperature from 1 to 16 months. There was a significant difference between exposure time and percent EI for *Cx. quinquefasciatus* (F = 30.6, df = 1, P < 0.001) with inhibition being significantly greater at 1, 2, 10, and 12 months compared with 3 and 16 months. We observed at the 3 month evaluation period that two replications may not have received the proper dose because no inhibition occurred.

An overall average of 67.2% EI was found for all species exposed to Altosid XR-G at 16 months continuous exposure to sunlight and ambient temperature. Generally, the EI of each species was significantly different than the control group (t = 4.5, df = 1, P = 0.02) with the exception of *Ae. albopictus*. Also no significant difference in EI occurred for *Ae. aegypti* and *Ae. taeniorhynchus* regardless of product exposure time.

Table 2 shows the EI of the three mosquito species treated with XR-G after one month exposure to two temperature regimes. There were no significant differences in EI within species to either temperature scenario (P > 0.05). Emergence inhibition was >75% after 1 month exposure for all three species regardless of temperature exposure. There was no emergence inhibition observed in the control group at either temperature regimes.

Previous studies have demonstrated excellent control of different mosquito species after application of sustained-release formulations of s-methoprene after multiple wetting events (Linthicum et al. 1989, Logan et al. 1990, Kramer and Beesley 1991, Qualls and Xue 2007). However, to our knowledge, this is the first study that has exposed such a product to extended environmental solar exposure without wetting. This information

Table 1. Percent emergence inhibition ± SE of four mosquito species treated with Altosid XR-G after solar exposure from 1 to 16 months.

Mosquito Species	Months					
	1	2	3	10	12	16
<i>Aedes aegypti</i>	*	*	*	85.7 ± 2.5 a	76.8 ± 4.5 a	78.9 ± 9.3 a
<i>Aedes albopictus</i>	*	*	*	82.1 ± 2.9 a	73.6 ± 4.5 a	58.9 ± 7.2 b
<i>Aedes taeniorhynchus</i>	*	*	*	89.4 ± 1.1 a	80.2 ± 3.3 a	76.2 ± 1.5 a
<i>Culex quinquefasciatus</i>	98.1 ± 0.5 a	95.9 ± 0.6 a	59.7 ± 5.9b	93.6 ± 1.1 a	83.6 ± 4.2 a	60.3 ± 2.6 b

*No data collected

*Means in rows with different letters represent statistically significant differences between months (P < 0.05).

Table 2. Mean percent emergence \pm SE of three mosquito species treated with Altosid XR-G after 1 month exposure to two different temperature scenarios.

Mosquito Species	15°C	39°C
<i>Aedes albopictus</i>	89.5 \pm 1.7	76.0 \pm 3.3
<i>Aedes aegypti</i>	80.6 \pm 3.5	77.9 \pm 3.7

No significant differences were observed between temperatures within each species (Student's t-test, $P > 0.05$).

is valuable for selecting whether or not application before a rain or tidal event is a viable option in areas where there is uncertainty regarding inundation. Our data showed that this product could be applied in the field for 10 months prior to a rain event and still result in significant mosquito emergence inhibition. In addition, the temperature scenario experiment provides further supporting evidence when using XR-G as a pre-treatment option. Our study demonstrated the efficacy and persistence of Altosid XR-G in the laboratory with potential implication of application in areas that become inaccessible following a flooding event. However, we do acknowledge that the efficacy against a field mosquito population still needs to be further evaluated.

IV. ACKNOWLEDGEMENTS

This research was supported by Wellmark International. This is a research report only. Anastasia Mosquito Control District does not endorse any of the products evaluated for mosquito control.

V. REFERENCES CITED

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EFFICACY OF GROUND ULV APPLICATION OF NYGUARD® (10% PYRIPROXYFEN) AGAINST AEADES ALBOPICTUS LARVAE IN ST. AUGUSTINE, FLORIDA

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ABSTRACT. Semi-field experiments were conducted on the efficacy of ground ultra-low volume (ULV) application of NyGuard Concentrate (10% pyriproxyfen) on adult emergence of *Ae. albopictus* larvae exposed as late third and early fourth instar. Direct contact was evaluated by direct exposure of pyriproxyfen to larvae in water collected when ULV conducted in the streets. Indirect contact was evaluated by removing treated vegetation from the streets and soaking the leaves in water for 24 hours before adding larvae in the laboratory. Both experiments indicated that ground ULV of NyGuard (10% pyriproxyfen) exhibited adult emergence inhibition >85%. Operationally, these results suggest that ground ULV application of 10% pyriproxyfen would suitably inhibit adult emergence of *Ae. albopictus*. We recommend that further testing be conducted so that this insect growth regulator can be labeled for area-wide ULV application as a larvicide.

Key Words: *Aedes albopictus*, pyriproxyfen, larvicide, emergence inhibition, vegetation

I. INTRODUCTION

Aedes albopictus (Skuse) is an imported and common anthropophilic container inhabiting mosquito (O'Meara 1997) that has been found to be a competent disease vector

in laboratory studies of more than 30 viruses including dengue. Because *Ae. albopictus* is a nuisance and competent vector of disease pathogens, control of this species is very important. One effective control method is to remove or empty water holding containers. However, for one reason or another, residents may be unwilling to remove or empty containers from their yards. If treatment with an insecticide is necessary, the abundance and sometimes small nature of individual containers impedes the ability of larvicides to reach every container, thus propagating mosquitoes. Treating containers using a ground ultra low volume (ULV) approach would not only cover more area but has the potential for better protection from nuisance and vector mosquitoes.

Through the years a wide range of larvicidal application methods against *Aedes spp.* have been evaluated (Ali et al. 1995, Sulaiman et al. 1997, Andrighetti et al. 2008, Gomez et al. 2011) including recent studies on the insect growth regulator (IGR) pyriproxyfen (Mulla 1991, Ali et al. 1995, Ali et al. 1997, Kono et al. 1997, Dhadialla et al. 1998, Sullivan 2000, Nayar et al. 2002, Chen et al. 2008, Invest and Lucas 2008). Currently, pyriproxyfen is not labeled for area-wide ULV application for mosquito control. However, this insecticide has been demonstrated to inhibit adult emergence of *Ae. aegypti* (L) after previous exposure as late third and early fourth instars at Anasta-

sia Mosquito Control District (AMCD), St. Johns County (SJC), Florida our mission is to protect the public from mosquito-borne disease. Detailed evaluation of new and advanced methods of control must be conducted in order to efficiently protect the people of SJC. Here we report on the efficacy of experimental ground ULV applications of NyGuard® (10% pyriproxyfen) to control *Ae. albopictus* by first exposing the larvae to this larvicide in field and laboratory studies.

II. MATERIALS AND METHODS

NyGuard® EC (AI 10% pyriproxyfen) was used in all studies and was provided by the Navy Entomology Center of Excellence (NECE), Jacksonville, FL. Because this product was not labeled for area-wide ULV application against mosquitoes in Florida, this experiment was conducted in tandem with NECE, who received an experimental use permit from the Florida Department of Agricultural and Consumer Services, to apply truck-mounted ULV applications of this chemical. All experiments were conducted in St. Augustine, Florida, during August and September 2012. The treatment and control areas each consisted of forty hectares of residential neighborhood. The control area was 3.22 km away from the treatment site. Both sites represented a typical neighborhood setting with similar vegetation where *Ae. albopictus* is often found by AMCD inspectors. Three houses, in each area, were chosen on separate streets as the sites for the experimental evaluations. Houses were >500 m from each other.

A Cougar truck mounted cold ULV aerosol sprayer (Clarke® Mosquito Control, Roselle, IL) was provided by NECE. Three applications of undiluted pyriproxyfen were conducted by a licensed AMCD applicator. Treatments occurred on 29 August, 7 and 25 September 2012. The first application was made at a rate of 319 ml/min and a speed of 13 km/hour. Two passes were made for the first trial. The second and third trials were dispensed as a single pass at a rate of 532 ml/min and a speed of 8.04 km/hr.

Direct Exposure Study A 946 ml plastic container (Ace Hardware, St. Augustine, FL.), filled with 200 ml of reverse osmosis tap water (GE SmartWater™ Reverse Osmosis Filtration System, Fairfield, CN) and placed at one (i.e. primary) residence and another cup directly across the street and labeled accordingly. Containers were placed in the vicinity of similarly leafed vegetation. Plant leaves were collected in zip lock bags at each site to provide an indication of the effectiveness of the active ingredient on vegetation when it rains as well as runoff into standing water. Though the plant type varied from each of the experimental sites, every effort was made to collect leaves of similar size and shape. No plants species names or leaf surface areas were notated during this experiment.

Indirect Exposure Study Ten leaves (average $2 \times 3 \text{ cm}^2$) from the treatment and 10 from control areas were removed and kept separately in zip lock bags and returned to the laboratory where they were immediately placed into 200 ml of reverse osmosis water in plastic containers. Leaves were soaked for 24 hours then removed from containers. Twelve cups were utilized for control and treated areas.

For both studies, immediately after application, treatment and control containers were brought back to the laboratory where ten late third/early fourth instar *Ae. albopictus* with average of 0.0749 g of food were added (Milk Bone™ Mini's Dog Treats, Del Monte Corporation). Larvae were previously reared from *Ae. albopictus* eggs obtained from the USDA-Agricultural Research Service, Center for Agricultural, Medical, and Veterinary Entomology, Gainesville FL in AMCD's insectary (insectary maintained at $25.5 \pm 0.5^\circ\text{C}$, 70-80% RH, and a 16L:8D photoperiod). Adult emergence data was collected for 2 weeks where larvae pupated and emerged, or died in the controls. Thus, adult emergence inhibition was calculated at the conclusion of the experiment, using the following equation:

$$\text{EI (\%)} = \frac{[A+D]}{\text{Total}} \times 100$$

(EI = emergence inhibition, A = alive larvae, D = dead larvae, Total = 10 larvae per container). Abbott's correction was

Table 1. Mean percent emergence inhibition for direct and indirect contact of three ULV application sites of pyriproxyfen. Location of placement data corresponds with primary and across the street locations for each application.

Placement Location	Direct Contact		Indirect Contact	
	Mean ± SEM ¹	P value ²	Mean ± SEM ¹	P value ²
836 W 2nd St	90 ± 10.00	0.71	90 ± 5.77	0.50
Across from: 836 W 2nd St	95 ± 05.00		95 ± 05.00	
824 W. 13th St	87 ± 13.33	0.42	90 ± 10.00	0.42
Across from: 824 W. 13th St	100 ± 0.00		100 ± 0.00	
894 South Volusia	77 ± 14.53	0.50	88 ± 06.23	0.20
Across from: 894 South Volusia	79 ± 01.50		97 ± 03.33	

¹Abbott's correction performed on data to account for emergence inhibition in controls.

²Paired t-test.

then performed on all data to account for emergence inhibition in controls (Abbott 1925). Paired t-tests were conducted on data to determine significant differences between location of containers (and associated vegetation) on each side of the street using GraphPad (GraphPad Software, Inc., La Jolla, CA.).

III. RESULTS AND DISCUSSION

At the primary and across the street locations no statistical difference in mean percent emergence inhibition occurred with

regard to direct treatment of containers (88.3%) or indirect contact (93.4%) in water from treated vegetation for each neighborhood (Table 1, Figures 1 and 2). The lack of statistical difference could be attributed to an even distribution of pyriproxyfen. However, we note that emergence inhibition was above 85% and may reflect a concentration-dependent factor of our application. Chism et al. (2003) demonstrated, under laboratory conditions, that the EI₉₅ for pyriproxyfen was 0.668 ppb, whereas our experiments applied undiluted 10% (AI) pyriproxyfen.

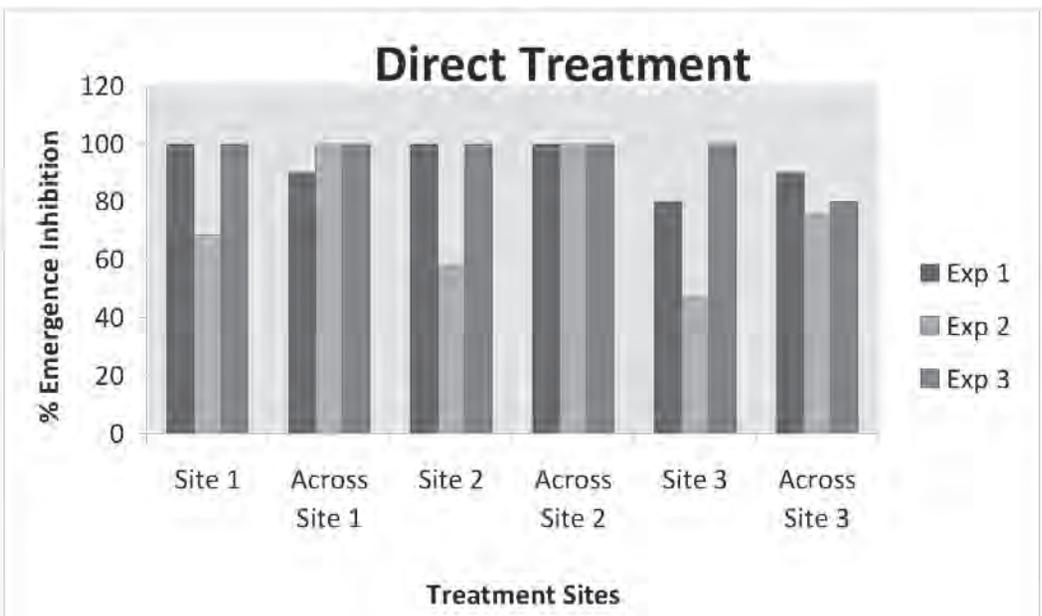


Figure 1. Mean percent emergence inhibition of adult *Ae. albopictus* (showing experimental variation) when directly exposed as 3rd-4th instar to water collected from the field where ground ULV applications of pyriproxyfen were conducted. The three sites coincide with the three houses chosen for the experiments. Abbott's correction performed on data to account for emergence inhibition in controls.

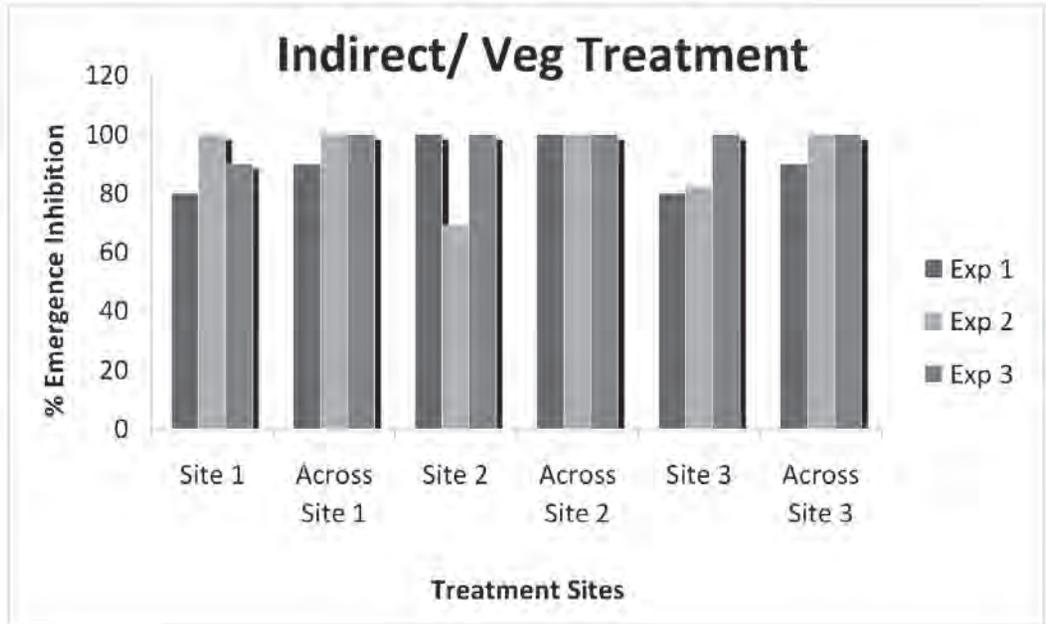


Figure 2. Mean percent emergence inhibition of adult *Ae. albopictus* (showing experimental variation) when exposed to ULV applications of pyriproxyfen as 3rd-4th instar indirectly in water from treated vegetation. The three sites coincide with the three houses chosen for the experiments. Abbott's correction performed on data to account for emergence inhibition in controls.

There are many studies that address the utilization of adult mosquitoes as a carrier of pyriproxyfen via horizontal transfer. Itoh et al. (1994) and Chism et al. (2003) exposed adult mosquitoes to surfaces that allowed them to pick up pyriproxyfen and horizontally transfer it to their next oviposition point. Later, Gaugler et al. (2012) proposed the use of auto dissemination stations that utilize the oviposition-seeking strategy of mosquitoes to disseminate pyriproxyfen into larval habitats. Although the study we conducted sought to discover the emergence inhibition efficacy of an ULV application of pyriproxyfen, there are strong implications that this application method could serve as a method to disperse this larvicide to larger areas than previously studied by adult transference. Our results strongly support that further research into ULV application of 10% pyriproxyfen for emergence inhibition of adult *Ae. albopictus* populations should be continued.

IV. ACKNOWLEDGMENTS

The authors would like to thank the US Navy Entomology Center of Excellence for

their assistance in this study. Further thanks go to Mike Smith and Patrick Kendrick for their valuable assistance in the field as well as Ali Fulcher and Richard Weaver for their help in the laboratory. This is a report on a research study and does not state, or imply, that the Anastasia Mosquito Control District of St. Johns County, Florida endorses any products.

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THE FIFTH ARBOVIRUS SURVEILLANCE & MOSQUITO CONTROL WORKSHOP

ANASTASIA MOSQUITO CONTROL DISTRICT (AMCD) OF ST. JOHNS COUNTY ST. AUGUSTINE, FLORIDA, MARCH 26-28, 2008

PROGRAM

Wednesday, March 26, 2008

Moderator: Dr. Rui-De Xue, Director, AMCD

- 8:50 am Welcome & Introduction: **Mrs. J. Moeller, AMCD Commissioner Chair, Dr. Gary Clark, Research Leader, USDA/ARS/CMAVE/MFRU, and Dr. Frank Van Essen, FMCA President**
- 9:00 am **Keynote Address:** West Nile Virus control in Sacramento and Yolo Counties, California **Mr. Dave Brown, Director of Sacramento-Yolo Vector Control District, Sacramento, CA**
- 9:30 am Affect of West Nile Virus on North American bird populations **Dr. John Edman, Emeritus Professor, University of California, Davis, CA**
- 10:10 am Present status of mosquito-borne diseases in India **Dr. B. Reddya Naik, Associate Professor, Osmania University, Hyderabad, India**
- 10:40 am Break
- 10:50 am Baits for mosquito control progress, an environmentally-friendly technique **Dr. Gunter Muller, Visiting Professor, The Hebrew University of Jerusalem, Israel**
- 11:20 am An overview of the Armed Forces Pest Management Board's programs **Dr. Stanton Cope, Captain, Medical Service Corps, US Navy Research Liaison Officer, Washington, DC**
- 11:50 am Florida Mosquito Control Association and its function **Dr. Frank Van Essen, President, FMCA, and Director, Collier Mosquito Control District, Naples, FL**
- 12:00 pm Lunch break (provided by Clarke Mosquito Control)

Moderator: Dr. Frank Van Essen, Director, Collier Mosquito Control District

- 1:00 pm An overview of USDA/ARS/CMAVE/Mosquito/Fly Research Unit programs **Dr. Gary Clark, Research Leader, USDA/ARS/CMAVE, Gainesville, FL**
- 1:30 pm A review of currently used surveillance techniques for *Aedes albopictus* (Skuse) **Dr. Dan Kline, Lead Scientist, Dr. Gary Clark, USDA/ARS/CMAVE, Gainesville, FL**
- 2:00 pm Topical and clothing repellent research at the MFRU **Dr. Uli Bernier, Research Chemist, USDA/ARS/CMAVE, Gainesville, FL**

- 2:30 pm Biting stable fly management **Dr. Jerry Hogsette, Research Entomologist, USDA/ARS/CMAVE, Gainesville, FL**
- 3:00 pm Break
- 3:10 pm Conducting adult mosquito control applications near organic operations **Mr. Gary Goodman, Assistant Director of Sacramento-Yolo Vector Control District, Sacramento, CA**
- 3:30 pm DNA Bar-coding: Molecular phylogenetics of mosquito biodiversity, **Dr. B. Reddy Naik, Associate Professor, Osmania University, Hyderabad, India**
- 3:50 pm Overview of UF Whitney laboratory research projects about vector mosquito larval physiology/molecular biology for possible control **Dr. Marco Neira, University of Florida, Whitney Laboratory, St. Augustine, FL**
- 4:30 pm End of session
- 6:30 pm Dinner and lecture: Overview of the DACS's Bureau of Entomology and Pest Control **Mr. Mike Page, Chief, Division of Agricultural Environments, DACS** Dinner location: Holiday Inn, A1A Beach Blvd, St. Augustine Beach, FL

Thursday, March 27, 2008

Moderator: Dr. Gary Clark, Research Leader, USDA/CMAVE

- 9:00 am Mosquitoes, and the pathogens that love them: An overview of the work of the new Emerging Pathogens Institute at the University of Florida **Dr. Glenn Morris, Director, Emerging Pathogens Institute, UF, Gainesville, FL**
- 9:30 am Capture rate responses of adult mosquitoes to light trap and human landing collection methods **Dr. Donald Barnard, Research Entomologist, Greg Knue, Dan. Kline, and Uli Bernier, USDA/CMAVE, Gainesville, FL**
- 9:50 am Mosquitoes make terrible neighbors: community outreach and education in Leon County, Florida **Dr. Richard Lobinske, Superintendent, and Judith Gross, Leon County Mosquito Control, Tallahassee, FL**
- 10:10 am Development of a high throughput mosquito assay for discovery of new toxicants for control **Dr. Jimmy Becnel, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 10:40 am Break
- 11:00 am Overview of mosquitoes and vector control programs in University of Florida's Urban Entomology Center, **Ms Alex Chaskopoulou and Dr. Phil Koehler, Professor, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 11:20 am Target site insensitivity-mediated pyrethroid resistance in *Culex quinquefasciatus* in Alabama **Dr. Nannan Liu, Associate Professor, Department of Entomology, Auburn University, AL**
- 11:40 am Sindbis virus: a probe to understanding the minutiae of mosquito biology **Dr. Doria Bowers, Associate Professor, Department of Biology, Univ. of North Florida, Jacksonville, FL**

- 12:00 pm ADAPCO updated technology **Mr. Derek Wright, ADAPCO, Sanford, FL**
 12:10 pm Lunch break (provided by ADAPCO)

Moderator: Dr. Dan. Kline, Lead Scientist, USDA/CMAVE

- 1:10 pm Florida Arbovirus Epidemiology, 2007 **Mrs. Rebecca Shultz, Arbovirus Coordinator, DOH, Tallahassee, FL**
- 1:30 pm An update of mosquitoes / disease surveillance and ongoing susceptibility testing to Dibrom in Jacksonville Mosquito Control **Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL**
- 1:45 pm Mosquito control surveillance in Volusia County **Mr. Jonas Stewart, Director, Volusia County Mosquito Control, Daytona Beach, FL**
- 2:00 pm Recent advancements in the Indian River Mosquito Control District's ground adulticiding program **Mr. John Plate, Indian River Mosquito Control District, Vero Beach, FL**
- 2:15 pm Break
- 2:30 pm Overview of Pasco County Mosquito Control District's program **Mr. Dennis Moore, Director, Pasco County Mosquito Control District, FL**
- 2:45 pm Update about Chatham County Mosquito Control programs **Mr. Robert Moulis, Entomologist, Chatham County Mosquito Control, Savannah, GA**
- 3:00 pm Overview of Collier County Mosquito Control District Programs **Dr. Frank Van Essen, Director, Collier Mosquito Control District, FL**
- 3:15 pm Vaportoxicity of 30 novel chemistries to *Aedes aegypti* and *Culex quinquesfasciatus* **Miss Alex Chaskopoulou, Department of Entomology & Nematology, University of Florida, Gainesville, FL**
- 3:30 pm Overview of AMCD's barrier treatments for mosquito control **Dr. Rui-De Xue, Director /Entomologist, AMCD, St. Augustine, FL**
- 3:40 pm Overview of the GTM NERR management plan, watershed implications **Dr. Michael Shirley, Director, Guana, Tolomato, Matanzas, National Estuarine Research Reserve, St. Augustine, FL**
- 3:55 pm Population dynamics of *Oc. infirmatus* and *Oc. atlanticus* in St. Johns County, Florida, 2004-2007 **Ms. Whitney Qualls, Biological Technician, Dr. Rui-De Xue, Director/Entomologist, AMCD, St. Augustine, FL**
- 4:10 pm End of session

Friday, March 28, 2008

Moderator: Dr. Harry Zhong, Professor, FAMU/PHEREC, Panama City, FL

- 9:00 am A review of new products for 2008 **Mr. David Sykes, B & G Chemical & Equipment Co.**
- 9:15 am Update about methoprene products for 2008 **Mr. Mel Whitson, Wellmark International, FL**
- 9:30 am An update on new product development from Clarke Mosquito Control **Mr. William Jany, Research Director, Clarke Mosquito Control**
- 9:45 am Pre-flood control of mosquitoes **Mr. Tom Floore, Research Entomologist, FAMU/PHEREC, Panama City, FL**

- 10:00 AMCD's aerial spraying contract service in 2007 **Mrs. Kay Gaines, Base Station Supervisor, AMCD, St. Augustine, FL**
- 10:15 am Update about nontarget study **Dr. Harry Zhong, FAMU, PHEREC, Panama City, FL**
- 10:30 am Keep moving forward **Mrs. Nicole Willams, Air Wolf Malaria Vector Research Institute, FL**
- 11:00 am **Break and laboratory tour/field trip**
- Moderator: Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL**
- 11:30 am Lunch and lecture: Overview of the University of Florida, Whitney Laboratory Programs **Dr. Peter Anderson, Professor/Director, University of Florida, Whitney Laboratory** (lunch provided by Central Life Sciences)
- 12:30 pm **Tour of the UF's Whitney Laboratory in St. Augustine, FL**
- 2:00 pm End of session and workshop

THE SIXTH ARBOVIRUS SURVEILLANCE & MOSQUITO CONTROL WORKSHOP & CELEBRATION OF AMCD'S 60 YEAR ANNIVERSARY

ANASTASIA MOSQUITO CONTROL DISTRICT (AMCD) OF ST. JOHNS COUNTY
ST. AUGUSTINE, FLORIDA, MARCH 31-APRIL 2, 2009

PROGRAM

Tuesday, March 31, 2009

Moderator: Dr. Rui-De Xue, Director/Entomologist, AMCD, St. Augustine, FL

- 8:50 am Welcome & Introduction **Ms. Janice Bequette, AMCD Commissioner Chairperson, and Dr. Ken Linthicum, Center Director, USDA/CMAVE/MFRU**
- 9:00 am **Keynote Address: Mosquito-borne arboviral diseases in the U.S.A. Dr. Lyle Petersen, Director, CDC, Division of Vector-Borne Infectious Diseases, Ft. Collins, CO**
- 9:40 am Rift Valley Fever Epidemics in Egypt: The Entomologic Investigations **Dr. Hanafi A. Hanafi, Head, Biological & Field Studies Section, Vector Biology Research Program, U.S. Naval Medical Research Unit No.3. Cairo, Egypt.**
- 10:00 am Insecticide-impregnated mosquito bed nets against mosquito-borne diseases in China **Dr. Tong-Yan Zhao, Professor & Director, Department of Vector Biology and Control, Institute of Microbiology and Epidemiology, Beijing, China**
- 10:25 am Impacts of insecticides on adult mosquito behaviors **Dr. Theeraphap Chareonviriyaphap, Professor, Department of Entomology, Kasetsart University, Bangkok, Thailand**
- 10:50 am Break
- 11:10 am Toxic baits for malaria vector mosquito control in African Countries **Mr. Amir Galili, Manager, Westham Innovations, Israel, Dr. Gunter Muller, Visiting Professor, The Hebrew University of Jerusalem, Israel**
- 11:30 am First 5 years of the DWFP Program to develop new and improved vector control methods and materials **Dr. G.B. White, Entomologist, University of Florida, Gainesville, FL**
- 11:50 am Update about the American Mosquito Control Association **Mr. Doug Carlson, President-Elect, AMCA**
- 12:05 pm Lunch break (provided by Clarke Mosquito Control)

Moderator: Dr. Gary Clark, Research Leader, USDA/CMAVE, Gainesville, FL

- 1:00 pm. Navy Entomology Center of Excellence and the Deployed War Fighter Protection Program **Dr. George Schoeler, Director, NECE, Jacksonville, FL**
- 1:20 pm Efficacy of commercial mosquito traps in capturing phlebotomine sand flies in Egypt **Dr. David Hoel, CDR, USDA/CMAVE, Gainesville, FL**
- 1:40 pm Environmental stewardship along Florida's Indian River Lagoon **Doug Carlson, Director, Indian River Mosquito Control District, Vero Beach, FL**
- 2:00 pm Biology of mosquito-borne arbovirus **Dr. Jonathan Day, Professor, University of Florida/IFAS, FMEL, Vero Beach, FL**
- 2:30 pm Effects of Sindbis virus variants on the mosquito host **Kristen A. Ciano and Dr. Doria F. Bowers, Associate Professor, Department of Biology, Univ. of North Florida, Jacksonville, FL**
- 3:00 pm Behaviors and response to repellents in Sindbis virus-infected mosquitoes **Whitney Qualls, Doria Bowers, Jonathan Day, and Rui-De Xue, AMCD, St. Augustine, FL**
- 3:15 pm Break
- 3:35 pm Host-seeking behaviors and trap development **Dr. Dan Kline, Lead Scientist, USDA/CMAVE, Gainesville, FL**
- 3:55 pm Research on topical repellents and repellent-treated uniform for the US military **Dr. Uli Bernier, Research Chemist, USDA/ARS/CMAVE, Gainesville, FL**
- 4:15 pm Finding the resting site of adult Phlebotomine sand flies in a village in Aswan, Egypt **Dr. Jerry Hogsette, Research Entomologist, USDA/ARS/CMAVE, Gainesville, FL**
- 4:35 pm Efficacy of ULV applications of *Bti* on container breeding mosquitoes **Dr. James C. Dunford, Medical Entomologist, Navy Entomology Center of Excellence, Jacksonville, FL**
- 4:55 pm Overview of UF Whitney laboratory research projects about vector mosquito larval physiology/molecular biology for possible control **Dr. Paul Linser, Professor, University of Florida, Whitney Laboratory, St. Augustine, FL**
- 5:15 pm End of session

Wednesday, April 1, 2009

Moderator: Dr. Dan Kline, Lead Scientist, USDA/CMAVE

- 8:40 am Temporal patterns of mosquito landing on human hosts: implications for vector detection, monitoring, and control **Dr. D.R. Barnard, G.J. Knue, C.Z. Dickerson, and R-D. Xue, USDA/CMAVE, Gainesville, FL**
- 9:00 am A review of colored lights used for mosquito trapping **Dr. Lee Cohnstaedt, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 9:20 am Traps for container-inhabiting mosquitoes **Mr. Peter Obenauer, Department of Entomology & Nematology, University of Florida, Gainesville, FL**
- 9:40 am Factors affecting foliar residues on vegetation **Dr. Sandra Allan, Research Entomologist, USDA/ARS/CMAVE, Gainesville, FL**

- 10:00 am The principles of pesticide application for barrier treatments: deposit distribution **Dr. Jane Bonds, Associate Professor, PHEREC/FAMU, Panama City, FL**
- 10:20 am Break
- 10:40 am Blood feeding response of mated and unmated nulliparous *Aedes aegypti* exposed to deltamethrin using an excito-repellent test system **Ms. Wasana Boonyuan, Department of Entomology, Kasetsart University, Bangkok, Thailand**
- 11:00 am Efficacy and non-target impacts of aerial application for adulticiding **Ms Alex Chaskopoulou, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 11:20 am Fly control research update **Mr. Joe Diclaro, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 11:40 am Evaluation of mosquito larvicides **Ryan Allen, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 12:00 pm Lunch break
- Moderator: Dr. Paul Linsler, Professor, UF/Whitney Lab, St. Augustine, FL**
- 1:00 pm Update about DACS' mosquito control programs **Mr. Mike Page, Chief, Entomology and Pest Control, DACS, Tallahassee, FL**
- 1:20 pm Arbovirus surveillance in LCMCD **Mr. James Burgess, Arbovirus Surveillance Coordinator, Lee County Mosquito Control District, Ft. Myers, FL**
- 1:40 pm Pesticide studies in Jacksonville Mosquito Control **Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL**
- 2:00 pm EEE surveillance in Volusia County **Mr. Jonas Stewart, Director, Volusia County Mosquito Control, Daytona Beach, FL**
- 2:20 pm Mosquito plant feeding behaviors & survival **Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL**
- 3:00 pm Break
- 3:20 pm AMCD's public and legislation education, **Mrs. Jeanne Moeller, Commissioner, AMCD, St. Augustine, FL**
- 3:40 pm Effect of DUET and its components on excitation behavior of *Culex quinquefasciatus* **Dr. Gary Clark, Research Leader, Dr. Sandra Allan, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 4:00 pm DUET laboratory and field evaluations and non-target impacts **Whitney Qualls, Rui-De Xue, Harry Zhong, and Bill Jany**
- 4:15 pm Effect of physiological conditioning on behavioral avoidance by using a single age group of *Aedes aegypti* exposed to deltamethrin and DDT **Ms. Suppaluck Polsomboon, Department of Entomology, Kasetsart University, Bangkok, Thailand**
- 4:30 pm Aerial application for mosquito control in St. Johns County **Mrs. Kay Gaines, Base Station Supervisor, AMCD, St. Augustine, FL**
- 5:55 pm Aerial ULV application of naled (2006-2008): impact on Miami blue butterfly larvae and efficacy against adult mosquitoes **Dr. Harry Zhong, Professor, FAMU, PHEREC, Panama City, FL**

- 5:10 pm End of session
- 6:30 pm Dinner (Holiday Inn, A1A Beach Blvd, St. Augustine Beach) with lecture: Overview of the GTM NERR program **Dr. Mike Shirley, Director, Guana, Tolomato, Matanzas, National Estuarine Research Reserve, St. Augustine, FL**

Thursday, April 2, 2009

Moderator: Dr. Harry Zhong, Professor, FAMU/PHEREC, Panama City, FL

- 8:50 am A new software and droplet size calibration for mosquito control operation **Mr. Bill Reynolds, Leading Edge Associations, LLC, NC.**
- 9:05 am A new adultciding on the markets **Mr. Mel Whitson, Regional Manager, Wellmark International, FL**
- 9:20 am Mosquito sentinel traps **Dr. Phil Kaufman, Assistant Professor, Department of Entomology & Nematology, UF, Gainesville, FL**
- 9:35 am ADAPCO updated technology in 2009 **Mr. Chris Peterson, ADAPCO, Sanford, FL**
- 9:50 am A review of new products **Mr. David Sykes, B & G Chemical & Equipment CO**
- 10:20 am Scientific free speech rights: conflict of interest, and their relation to pesticide use **Mr. Ed. Slavin, St. Augustine, FL**
- 10:35 am Workshop end and 60-year celebration activity
- 10:40-11:30 am Tour of AMCD posters, lab, equipment, and facility at the Base Station
- 11:30 am-
12:10 pm Ceremony (**Moderator: Ms. Janice Bequette, Chairperson of the Board of Commissioners**)
- 12:10-1:30 pm Lunch provided and end of workshop.

THE SEVENTH ARBOVIRUS SURVEILLANCE & MOSQUITO CONTROL WORKSHOP

ANASTASIA MOSQUITO CONTROL DISTRICT (AMCD) OF ST. JOHNS COUNTY ST. AUGUSTINE, FLORIDA, MARCH 23-25, 2010

PROGRAM

Tuesday, March 23, 2010

Moderator: Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL

- 8:50 am Welcome & Introduction **Mrs. Jeanne Moeller, AMCD Commissioner Chairperson and Dr. Gary Clark, Research Leader, USDA/CMAVE/MFRU**
- 9:00 am **Keynote Address:** Control of mosquito-borne diseases: future direction **Dr. Kenneth Linthicum, Center Director, USDA/ARS/CMAVE, Gainesville, FL**
- 9:50 am Arthropod-borne virus in China **Dr. Guo-Dong Liang, Professor and Deputy Director, Institute for Viral Disease Prevention and Control, China CDC, Beijing, China**
- 10:20 am Sugar toxic baits for mosquito and sand fly control **Mr. Amir Galili, Manager, Westham Innovations, Israel and Dr. Gunter Muller, Visiting Professor, The Hebrew University of Jerusalem, Israel**
- 10:40 am Break
- 10:55 am How can we counter the bio-terrorist threat? **Mr. Mark Fedders, FBI Supervisory Special Agent, Washington, DC**
- 11:20 am Bioterrorism by pathogen-carrying mosquitoes – a threat or hoax? **Dr. William R. Harvey, Professor, UF Whitney Laboratory, St. Augustine, FL**
- 11:40 am Overview of the American Mosquito Control Association. **Mr. Doug Carlson, President, AMCA**
- 12:00 pm Lunch break (provided by ADAPCO)

Moderator: Dr. Gary Clark, Research Leader, USDA/CMAVE, Gainesville, FL

- 1:00 pm Epidemiology of EEE and surveillance of vector mosquitoes in Florida **Dr. Jonathan Day, Professor, University of Florida, FMEL, Vero Beach, FL**
- 1:20 pm Environmental stewardship along Florida's Indian River Lagoon **Doug Carlson, Director, Indian River Mosquito Control District, Vero Beach, FL**

- 1:40 pm Overview of Florida Mosquito Control Association **Mrs. Shelly Redovan, President, FMCA and Deputy Director of Lee County Mosquito Control District, FL**
- 2:00 pm Update about USDA's area-wide project for control of *Aedes albopictus* **Dr. Gary Clark, Research Leader, USDA/ARS/CMAVE, Gainesville, FL**
- 2:20 pm Mosquito population surveillance system in China **Dr. Qiyong Liu, Professor and Director, Department of Vector Biology and Control, Assistant Director, Institute for Communicable Disease prevention and Control, China CDC, Beijing, China**
- 2:40 pm Break
- 3:00 pm Update on mosquito control traps and trapping **Dr. Dan Kline, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 3:20 pm Impact of vegetation on ULV spray and barrier effectiveness **Dr. Jane Bonds, Associate Professor, FAMU/PHEREC, Panama City, FL**
- 3:40 pm Influencing factors and improvement for aerial spraying efficacy **Mr. Mark Latham, Director, Manatee County Mosquito Control District, FL**
- 4:00 pm Non-target impacts of mosquito adulticiding **Ms Alex Chaskopoulou, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 4:20 pm Insecticide resistance in Florida house flies **Dr. Phil Kaufman, Assistant Professor, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 4:40 pm Novel method of controlling flies **Mr. Joe Diclaro and Dr. Phil Koehler, Professor, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 5:00 pm End of session
- 5:00-5:30 pm Committee meeting for the 2nd International Forum for Surveillance and Control of Mosquitoes and Mosquito-borne Diseases, Beijing, May 23-27, 2011 (**Dr. Tong-Yan Zhao and Dr. Rui-De Xue**)
- 6:30 pm Dinner and lecture (Holiday Inn, A1A Beach Blvd., St. Augustine Beach) Overview of aerial capability in Florida mosquito control organizations **Mr. Mark Latham, Director, Manatee County Mosquito Control District, FL.**

Wednesday, March 24, 2010

Moderator: Dr. Dan Kline, Research Entomologist, USDA/CMAVE

- 8:40 am Mosquito attractants and repellents **Dr. Uli Bernier, Research Chemist, USDA/CMAVE, Gainesville, FL**
- 9:00 am Mosquito magnets with attractant study in China **Dr. Tong-Yan Zhao, Professor & Director, Beijing Inst of Microbiology and Epidemiology, Beijing, China**
- 9:20 am Do I stay or do I go? Predicting mosquito behavior in the presence of repellents, insecticides, and attractants **Dr. Lee Cohnstaedt, Research Entomologist, USDA /ARS /CMAVE, Gainesville, FL**

- 9:40 am Operational entomology in the Navy Entomology Center of Excellence
Dr. Craig Stoops, Navy Entomology Center of Excellence, Naval Air Station, Jacksonville, FL
- 10:00 am Jacksonville Mosquito Control's aerial program and aerial spray in 2009
Mr. Richard Smith, Superintendent, Jacksonville, FL
- 10:20 am Unusual population increase and aerial spraying in St. Johns County in 2009
Dr. Rui-De Xue, Director, and Mrs. Kay Gaines, Base Station Supervisor, AMCD, St. Augustine, FL
- 10:20 am Break
- 10:30 am Mosquito larval physiology/molecular biology and possible use for control
Dr. Paul Linser, Professor, University of Florida, Whitney Laboratory, St. Augustine, FL
- 10:50 am Source reduction and coastal wetland restoration in NE Florida
Mr. Ron Brockmeyer, Environmental Scientist, St. Johns Water Management District, Palatka, FL
- 11:10 am New innovations in biocontrol of mosquitoes
Dr. Jimmy Becnel, Research Entomologist, USDA/ARS/CMAVE, Gainesville, FL
- 11:30 am Permits for exotic mosquito research in Florida
Dr. Peter Jiang, Entomologist, DACS, Tallahassee, FL
- 11:50 am Update about EPA/AMCA pesticide environmental stewardship program
Mr. Doug Wassmer, Entomologist, Pasco County Mosquito Control District, FL
- 12:10 pm Lunch break (provided by Central Life Sciences)
- Moderator: Dr. Paul Linser, Professor, UF/Whitney Lab, St. Augustine, FL**
- 1:00 pm Update about DACS' mosquito control programs & regulation
Mr. Mike Page, Chief, Entomology and Pest Control, DACS, Tallahassee, FL
- 1:20 pm Arbovirus surveillance report in Florida, 2009
Ms. Elizabeth Radke, Arbovirus Coordinator, DOH, Tallahassee, FL
- 1:40 pm Population surveillance and research study in Jacksonville Mosquito Control
Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL
- 2:00 pm Operation programs in Volusia County, 2009
Mr. Jonas Stewart, Director, Volusia CMC, Daytona Beach, FL
- 2:20 pm What surprised mosquito control in St. Johns County, 2009?
Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL
- 2:40 pm Overview of Beach Mosquito Control District programs
Mr. James Clauson, Director, Beach Mosquito Control District, Panama City, FL
- 3:00 pm Break
- 3:15 pm Achievements of AMCD's education/public relation program in 2009
Mr. Adam Holt, Education Specialist, AMCD, St. Augustine, FL
- 3:30 pm Enhancing mosquito control awareness in Hernando County, Florida
Dr. Guang-Ye Hu, Director, Hernando County Mosquito Control, FL
- 3:45 pm Marsh restoration and its impact on mosquito populations
Mr. Andrew Thornton, Field Biologist, AMCD/DEP, St. Augustine, FL

- 4:00 pm Impacts of barrier spraying on honey bees **Whitney Qualls, Biologist, Rui-De Xue (AMCD, St. Augustine, FL) and Harry Zhong (FAMU/ PHEREC, Panama City, FL)**
- 4:15 pm Impact of lake and aquatic weed control on mosquitoes and midges **Mr. Stu Metzler, Aquagenix/DBI, Jacksonville, FL**
- 4:30 pm House fly dispersal from, and *Escherichia coli* 0157:H7 prevalence, on dairy farms in north central Florida **Ms. Roxanne Burrus, USDA/ CMAVE, Gainesville, FL**
- 4:50 pm Update on blue-black cloth targets for stable fly management: size matters, or does it? **Dr. Jerry Hogsette, Research Entomologist, USDA/ CMAVE, Gainesville, FL**
- 5:10 pm End of session

Thursday, March 25, 2010

Moderator: Dr. Harry Zhong, Professor, FAMU/PHEREC, Panama City, FL

- 8:50 am New droplet size collection and calibration for mosquito control operation **Mr. Bill Reynolds & Mr. Mike Reynolds, Leading Edge Associations, LLC, NC**
- 9:10 am Update-new products for mosquito control for 2010 **Mr. Mel Whitson, Regional Manager, Wellmark International, FL**
- 9:35 am ADAPCO updated technology in 2010 **Mr. Chris Peterson, ADAPCO, Sanford, FL**
- 9:50 am Online database for mosquito and disease surveillance **Dr. Harry Zhong, Professor, FAMU/PHEREC, Panama City, FL**
- 10:10 am Break
- 10:25 am Natular™ stewardship initiatives over the past two seasons **Mr. Jim McNelly, Director of Environmental Sciences and Griffith Lizarraga, Field Biologist, Clarke Technical Center, Schaumburg, IL**
- 10:40 am Do I stay or do I go? Predicting mosquito behavior in the presence of repellents, insecticides, and attractants **Dr. Lee Cohnstaedt, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 11:00 am Update on non-target studies at FAMU/PHEREC **Dr. Harry Zhong, Professor, FAMU, PHEREC, Panama City, FL**
- 11:20 am DACS's inspection report **Mr. Steven Harrison, Environmental Manager, DACS, Tallahassee, FL**
- 11:40 am Update about MapVison in AMCD's operation **Mr. Richard Weaver, Data Manager, AMCD, St. Augustine, FL**
- 12:00 pm Lunch break (provided by Clarke Mosquito Control) and workshop end
- 12:30 pm AMCD open house, facility tour, and public meeting about mosquito control (AMCD Board Room). All welcome to attend.
- 1:00 pm Special Presentation: Home and Garden: Mosquito Control Prevention (AMCD Board room).. **Keith Fuller, UF Extension Agent, St. Augustine, FL**

THE EIGHTH ARBOVIRUS SURVEILLANCE & MOSQUITO CONTROL WORKSHOP

ANASTASIA MOSQUITO CONTROL DISTRICT (AMCD) OF ST. JOHNS COUNTY ST. AUGUSTINE, FLORIDA, MARCH 29-31, 2011

PROGRAM

Tuesday, March 29, 2011

Panel Session: Moderator: Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL

- 8:50 am Welcome & Introduction **Mr. Ron Radford, AMCD Commissioner Chair, and Dr. Gary Clark, Research Leader, USDA/CMAVE/MFRU**
- 9:00 am **Keynote Speaker:** The national needs and future direction for controlling mosquitoes and mosquito-borne diseases **Dr. Daniel Strickman, National Program Leader, USDA/ARS, Beltsville, MD**
- 9:50 am **Guest Speaker:** Dengue vector control and insecticide resistance in *Aedes aegypti* in Taiwan **Dr. Err-Lieh Hsu, Emeritus Professor, Department of Entomology, National Taiwan University, Taipei, Taiwan**
- 10:20 am Chickungunya outbreak and its management in southern China **Dr. Tong-Yan Zhao, Professor & Director, Dept. of Vector Biology and Control, Beijing Institute of Microbiology and Epidemiology, Beijing, China**
- 10:40 am Break
- 10:50 am Risk assessments of mosquito-borne diseases in Florida **Dr. Jonathan F. Day, Professor, University of Florida, FMEL, Vero Beach, FL**
- 11:20 am Sugar toxic baits for sand fly control in Israel and Africa **Dr. Gunter Muller, Visiting Professor, The Hebrew University of Jerusalem, Israel**
- 11:40 am Blood sucking bugs - Bedbugs bite back and their management **Dr. Roberto Pereira, Associate Research Scientist, Department of Entomology & Nematology, UF, Gainesville, FL**
- 12:00 pm Lunch break (provided by Clarke Mosquito Control)

Other Projects Session:

Moderator: Dr. Dan Kline, Research Entomologist, USDA/CMAVE, Gainesville, FL

- 1:00 pm Addressing the challenge of mangrove mosquitoes: Planning modification of larval habitats integrating diverse biophysical data in Australia **Dr. Pat Dale, Professor, Jon Knight, and Clive Easton, Griffith University, Australia**
- 1:20 pm A history of Florida's Subcommittee on Managed Marshes: Controlling mosquitoes while enhancing the environment **Mr. Doug Carlson, Director, Indian River Mosquito Control District, Vero Beach, FL**

- 1:40 pm Overview of the American Mosquito Control Association **Dr. Roxanne Connelly, Vice President, AMCA and Associate Professor, UF /FMEL, Vero Beach, FL**
- 1:50 pm Overview of the Florida Mosquito Control Association **Dr. Roxanne Connelly, President, FMCA and Associate Professor, UF/FMEL, Vero Beach, FL**
- 2:00 pm A mosquito control surveillance system using sub-meter satellite imagery and location-based models **Dr. Robert Novak, Professor, University of Alabama, Birmingham, AL**
- 2:20 pm West Nile outbreak and control measurement in Greece **Dr. Alex Chaskopoulou, Post Doctor, USDA Lab, Greece and Dr. Phil Koehler, Professor, Department of Entomology & Nematology, Univ. of Florida, Gainesville, FL**
- 2:40 pm Break
- 2:50 pm Novel method of controlling flies **Dr. Joe Diclaro and Dr. Phil Koehler, Professor, Department of Entomology & Nematology, UF, Gainesville, FL**
- 3:10 pm House flies: Specific traps for specific location on farms **Dr. Jerry Hogsette, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 3: 25 pm Update on the Deployed War-Fight Protection Program on R & D of new methods and materials for vector control and personal protection by the U.S. Armed Forces Pest Management Board **Lt. Col. Dr. Douglas Burkett, AFPMB and Dr. Graham White, University of Florida/IFAS, Department of Entomology and Nematology, Gainesville, FL**
- Dengue Fever Session:*
- 3:40 pm Experience with eradication of *Aedes aegypti* in the Americas and how it could relate to the Florida Keys **Dr. Gary Clark, Research Leader, USDA/CMAVE, Gainesville, FL**
- 4:00 pm The model concepts and its application for dengue fever vector surveillance and management **Dr. Dana Focks, Research Professor, UF/ Emerging Pathogens Institute, Gainesville, FL**
- 4:20 pm The dengue fever vector and its insecticide resistance in China **Dr. Feng-Xia Meng, Professor, Department of Vector Biology and Control, Institute for Communicable Disease prevention and Control, China CDC, Beijing, China**
- 4:40 pm Arbovirus surveillance and dengue epidemic in Florida, 2010 **Dr. D. Stanek, Epidemiologist, DOH, Tallahassee, FL**
- 5:00 pm End of workshop session
- 5:20-5:40 pm 2nd Committee meeting for the 2nd International Forum for Surveillance and Control of Mosquitoes and Mosquito-Borne Diseases, Beijing, May 23-27, 2011 **Dr. Tong-Yan Zhao and Dr. Rui-De Xue**
- 6:30 pm Dinner & lecture: (Holiday Inn, A1A Beach Blvd, St. Augustine Beach) Overview of malaria control projects in Africa **Dr. Gunter Muller, Visiting Professor, The Hebrew University of Jerusalem, Israel**

Wednesday (March 30, 2011)**Repellent /Attractant Session:****Moderator: Dr. Gary Clark, Research Leader, USDA/CMAVE, Gainesville, FL**

- 8:40 am Evaluation of spatial repellents for mosquitoes and fly management systems **Dr. Dan. Kline, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 9:00 am Field evaluation of spatial repellents against *Aedes albopictus* and *Aedes taeniorhynchus* **Dr. Rui-De Xue, Whitney Qualls, Mike Smith, Kay Gaines, Richard Weaver, AMCD, St. Augustine, FL**
- 9:10 am Response of Sindbis virus-infected *Aedes aegypti* to natural repellents and DEET **Whitney Qualls, Biologist, AMCD, St. Augustine, FL**
- 9:30 am Efficacy testing of repellent compounds using cloth and membrane in vitro screening methods **Ms. Natasha Elejalde, USDA/CMAVE, Gainesville, FL**
- 9:50 am Modeling, synthesis, and bioassay of new public health insecticides **Dr. Maia Tsikolia, Research Chemist, USDA/CMAVE, Gainesville, FL**
- 10:10 am Field evaluation of B & G sentinel traps with 3 attractants against *Aedes albopictus* in, Elkton, Florida **Mr. Mike Smith, Biotech, AMCD, St. Augustine, FL**
- 10:20 am Break

Larval/Adult Control Session:

- 10:35 am Update research on mosquito larval physiology and possible use for control **Dr. Paul Linser, Professor, University of Florida, Whitney Laboratory, St. Augustine, FL**
- 10:55 am Effect of Natular (spinosad) applied to catch basins breeding *Culex pipiens quinquefasciatus* mosquitoes in a Jacksonville subdivision **Mr. Richard Smith, Superintendent, Jacksonville Mosquito Control Division, Jacksonville, FL**
- 11:05 am Update about microbial larvicides for mosquito biocontrol **Mr. Frank Clarke, Clarke Mosquito Control, Kissimmee, FL**
- 11:20 am Field evaluation of ULV spray, lethal ovitrap, and backyard spray for control of dengue fever vector, *Aedes albopictus* **Dr. Chun-Xiao Li, Associate Professor, Beijing Institute of Microbiology and Epidemiology, Beijing, China**
- 11:35 am Ground application of Aquesolin and Aqua Kontrol against *Anopheles quadrimaculatus*, *Aedes aegypti*, and *Aedes albopictus* **Dr. Rui-De Xue, AMCD, St. Augustine, FL and Dr. James Brown, Paint Rock River Consulting, Gurley, AL**
- 11:50 am ULV and thermal fog applications against mosquitoes and sand flies in hot-arid environments **Dr. Seth Britch, Entomologist, Dr. Ken Linthicum, Center Director, USDA/CMAVE, Gainesville, FL**
- 12:05 pm Lunch Break (provided by the Central Life Sciences)

Program & Legislation Session:**Moderator: Dr. Paul Linser, Professor, UF/Whitney Lab, St. Augustine, FL**

- 1:00 pm Update about entomological research projects in the Navy Entomology Center of Excellence **Dr. Craig Stoops, Navy Entomology Center of Excellence, Naval Air Station, Jacksonville, FL**

- 1:20 pm Overview of the AMCA's legislation and NPDES **Mr. Joseph Conlon, Technical Advisor, American Mosquito Control Association, Mount Laurel, NJ**
- 1:40 pm Update on DACS' mosquito control regulatory activities and NPDES **Mr. Mike Page, Chief, Entomology and Pest Control, DACS, Tallahassee, FL**
- 2:00 pm How to prevent bioterrorist's threats **Mr. Will So, Policy Program Specialist and Ms. Carla Lee, FBI Agent, FBI Biological Countermeasures, Jacksonville, FL**
- 2:20 pm Arbovirus surveillance program and the first malaria case in Jacksonville in 2010 **Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL**
- 2:40 pm Aquatic invertebrates and mosquito population surveillance in Guana **Mr. Andrew Thornton, Field Biologist, AMCD/DEP, St. Augustine, FL**
- 3:00 pm Break
- 3:30 pm Efficacy of ovitrap colors and patterns for attracting *Aedes albopictus* at suburban field sites in north central Florida **Dr. David Hoel, Entomologist, U.S. Navy**
- 3:50 pm Overview of Lee County Mosquito Control Programs in 2010 **Mrs. Shelly Redovan, Deputy Director, Lee County Mosquito Control District, Ft. Myers, FL**
- 4:10 pm Overview of Volusia Mosquito Control Programs in 2010 **Mr. Bruce Morgan, Supervisor III, Volusia Mosquito Control District, New Smyrna Beach, FL**
- 4:30 pm Overview of Beach Mosquito Control District programs in 2010 **Mr. James Clauson, Director, Beach Mosquito Control District, Panama City, FL**
- 4:50 pm What have we done in 2010? Overview of programs **Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL**
- 5:05 pm End of session

Thursday, March 31, 2011

Technology and Non-target Session:

Moderator: Dr. Harry Zhong, Professor, FAMU/PHEREC, Panama City, FL

- 9:00 am MapVision/VCMS applications at AMCD **Mr. Richard Weaver, Data Manager, AMCD, St. Augustine, FL**
- 9:20 am Update on new products for mosquito control for 2011 **Mr. Mel Whitson, Regional Manager, Wellmark International, Melbourne, FL (merged to other talk)**
- 9:40 am ADAPCO updated technology for 2011 **Mr. Chris Peterson, ADAPCO, Sanford, FL**
- 10:00 am The LongRay fogger machine's overview **Mr. Brian Liang, LongRay Manufacturer, Shenzhen, China**
- 10:20 am Overview of Clarke's new products for 2011 **Mr. Frank Clarke, Clarke Mosquito Control, Kissimmee, FL**

- 10:40 am Mosquito larval control: a new formulation, **Mr. John Olsen, President, Cree Industries, Jacksonville, FL**
- 11:00 am Update about new biocontrol agents and its non-target impacts **Dr. Harry Zhong, Professor, FAMU, PHEREC, Panama City, FL**
- 11:20 am MADIS techniques for mosquito control operation & program management **Mr. John Corbett and Dr. Robert Novak, University of Alabama, Birmingham, AL**
- 11:40 am Products on the market from UNIVAR for 2011 **Mr. John Ryan, Public Health Industry Specialist, Univar USA, Inc., Orlando, FL**
- 11:50 am Scientific free speech rights **Mr. Ed. Slavin, St. Augustine, FL**
- 12:00 pm Lunch break (provided by ADAPCO) and end of workshop

THE NINTH ARBOVIRUS SURVEILLANCE AND MOSQUITO CONTROL WORKSHOP

ANASTASIA MOSQUITO CONTROL DISTRICT (AMCD) OF ST. JOHNS COUNTY ST. AUGUSTINE, FLORIDA, MARCH 27-29, 2012

PROGRAM

Tuesday, March 27, 2012

Panel Session:

Moderator: Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL

- 8:30 am Welcome & Introduction **Mrs. Jeanne Moeller, AMCD Commissioner Vice Chairperson, Dr. Kenneth Linthicum, Center Director, USDA/CMAVE, and Mr. Robert Betts, FMCA President-Elect**
- 8:40 am Keynote Speaker: The research program of the CDC's Entomological Branch. **Dr. Robert Wirtz, Branch Chief, CDC, Atlanta, GA**
- 9:30 am Guest Speaker: Overview of attractive toxic sugar baits against mosquitoes **Dr. Gunter Müller, Visiting Scientist, The Hebrew University of Jerusalem, Israel**
- 9:50 am DNA bar-coding techniques for mosquito systematics **Dr. Tong-Yan Zhao, Professor & Director, Dept. of Vector Biology and Control, Beijing Institute of Microbiology and Epidemiology, Beijing, China**
- 10:10 am Genotyping of *Aedes albopictus* species in several Florida counties **Dr. Azliyati Azizan, Assistant Professor, Dept. of Global Health, University of South Florida, Tampa, FL**
- 10:30 am Break

Moderator: Dr. Gunter Müller, Visiting Professor, The Hebrew University of Jerusalem, Israel

- 10:45 am Mosquitoes and mosquito-borne disease surveillance and control in Liberia. **Dr. Peter Obenauer, Entomologist, Navy Entomology Center for Excellence, Jacksonville, FL**
- 11:05 am Overview of "The International Forum for Surveillance and Control of Mosquitoes and Mosquito-borne Diseases" **Dr. Tong-Yan Zhao and Dr. Rui-De Xue, Co-organizers for the International Forum**
- 11:20 am Overview of the Society of Vector Ecology. **Dr. Ken Linthicum, President, SOVE and Center Director, USDA/CMAVE, Gainesville, FL**
- 11:40 am Overview of the Florida Mosquito Control Association **Mr. Robert Betts, President-Elect, FMCA and Director, Escambia County Mosquito Control, FL**
- 12:00 pm Lunch break (provided by Clarke Mosquito Control)

Arbovirus Session:**Moderator: Dr. Gary Clark, Research Leader, USDA/CMAVE, Gainesville, FL**

- 1:00 pm Risk assessments of arbovirus through bird breeding survey and model. **Dr. Jonathan F. Day, Professor, University of Florida, FMEL, Vero Beach, FL**
- 1:20 pm EEE vector mosquito behavior and ecological habitats **Dr. Nathan Burket-Cadena, University of South Florida, Tampa, FL**
- 1:40 pm Multiple dengue introductions in Florida **Dr. Danielle Stanek, Epidemiologist, DOH, Tallahassee, FL**
- 2:00 pm WNV outbreak in Jacksonville, Florida, 2011 **Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL**
- 2:15 pm Jacksonville Mosquito Control Response for WNV outbreak **Mr. Richard Smith, Superintendent, Jacksonville Mosquito Control, FL** (presented by Ms. Marah Clark)
- 2:30 pm Break

Malaria Control Session:**Moderator: Dr. Robert Novak, Professor, University of South Florida, Tampa, FL**

- 2:50 pm Behavioral ecology of malaria vector mosquitoes in Southern Zambia **Dr. Douglas Norris, Associate Professor, Bloomberg School of Public Health, Johns Hopkins University, Baltimore, MD**
- 3:10 pm Evaluation of mosquito coils against *Aedes aegypti*, *Anopheles albimanus*, and *Culex quinquefasciatus* **Dr. Rui-De Xue, W. Qualls, J. Phillips, AMCD, St. Augustine, FL**
- 3:30 pm How much vector control is needed to achieve malaria elimination? **Dr. John Beier, Professor, University of Miami, Miami, FL**
- 3:50 pm Malaria control in Uganda **Dr. Robert Novak, Professor, University of South Florida, Tampa, FL**

Host /Mosquitoes & Pathogen Interaction Session:**Moderator: Dr. Seth C. Britch, Research Entomologist, USDA/ARS/CMAVE, Gainesville, FL**

- 4:10 pm Vector competence of Florida mosquitoes for Rift Valley fever virus **Michael J. Turell, David J. Dohm, US Army Medical Research Institute for Infectious Diseases; Kenneth J. Linthicum, Robert L. Aldridge, Seth C. Britch, USDA-ARS Center for Medical, Agricultural, & Veterinary Entomology; Carl Boohene, Nick Harboe, Michael J. Mahler, Polk County Mosquito Control** (presented by Seth C. Britch)
- 4:30 pm Sindbis virus infection and its impacts on blood feeding behavior & repellency by *Aedes aegypti* **Dr. Whitney Qualls, Biologist, AMCD, St. Augustine, FL**
- 4:50 pm Frog-biting mosquitoes of Florida **Mr. Eric M. Blosser and Dr. Phil Lounibos, Florida Medical Entomology Laboratory, University of Florida, Vero Beach, FL**
- 5:10 pm End of session

- 5:10-5:40 pm. 3rd committee meeting for the 3rd International Forum for Surveillance and Control of Mosquitoes and Mosquito-Borne Diseases, Suzhou, May 27-31, 2013 **Dr. Tong-Yan Zhao & Dr. Rui-De Xue**
- 6:30 pm Dinner and lecture (Holiday Inn, A1A Beach Blvd, St. Augustine Beach) Overview of Whitney Laboratory projects **Dr. Peter Anderson, Director and Professor, University of Florida, Whitney Laboratory**

Wednesday, March 28, 2012

Behaviors, Repellent /Attractant Session:

Moderator: Dr. Dan Kline, Research Entomologist, USDA/CMAVE, Gainesville, FL

- 8:30 am Mosquito bite protection, performance of permethrin-treated US military uniforms **Dr. Ulrich R. Bernier, Research Chemist, USDA/CMAVE, Gainesville, FL**
- 8:50 am Trapping Tabanidae at the Lower Suwannee Wildlife Refuge located in Levy & Dixie Counties, Florida. **Dr. Dan. Kline, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 9:10 am Sand fly control through ATSB **Dr. Gunter Müller, Visiting Professor, The Hebrew University of Jerusalem, Israel**
- 9:20 am Examining mosquito repellent dose-response using a module on human skin and human-worn silicone membranes **Ms. Natasha Elejalde, Dr. Ulrich R. Bernier, Research Chemist, USDA/CMAVE, Gainesville, FL**
- 9:35 am *Aedes albopictus* oviposition preference for different sized containers and availability of flowering *Buddleja davidii* plants **Tim Davis, UF/Dept of Entomology and Nematology, Dan Kline, USDA/CMAVE, Phil Kaufman, UF/Dept. of Entomology and Nematology, Jerry Hogsette, USDA/CMAVE, Andy Tatem, UF/Emerging Pathogens Institute & Dept. of Geography, Gainesville, FL**
- 9:55 am Key factors in barrier trapping: model studies **Dr. C. Lord, Associate Professor, University of Florida, Vero Beach, FL**
- 10:15 am Field efficacy of mosquitito and BG-Sentinel traps for the collection of *Aedes albopictus* in Leon County, North Florida **Dr. Peter Jiang, Entomologist, Bureau of Pesticides, DACS, Tallahassee, FL**
- 10:30 am Break

Larval/Adult Control & Resistance Session:

Moderator: Dr. Jerry Hogsette, Research Entomologist, USDA/CMAVE, Gainesville, FL

- 10:45 am New model of action of insecticides **Dr. Jeff Bloomquist, Professor, Emerging Pathogen Institute, University of Florida, Gainesville, FL**
- 11:05 am Insecticidal activity of trifluoromethylphenyl carboxamides against mosquitoes. **Dr. Maia Tsikolia, Research Chemist, USDA/CMAVE, Gainesville, FL**
- 11: 20 am Potential of using insecticidal paint for controlling mosquitoes **Ephraim Ragasa and Phil Koehler, University of Florida, Dept. of Entomology and Nematology, Gainesville, FL**
- 11:35 am Insecticide resistance surveillance of vector mosquitoes in Jiangsu, China **Mrs. Wei-Fang Yang, Associate Professor, Dr. Yan Xu, Director,**

Department of Vector Control and Disinfection, Jiangsu CDC, Nanjing, China

- 11:50 am Yeast particle encapsulation as a potential solution to pesticide resistance **Dr. Gary Ostroff, Research Professor, University of Massachusetts Medical School, Worcester, MA**
- 12:10 pm Lunch break (provided by Central Life Sciences)

Program & Legislation Session:

Moderator: Dr. Ulrich R. Bernier, Research Chemist, USDA/CMAVE, Gainesville, FL

- 1:10 pm Florida mosquito control history **Dr. Gordon Patterson, Professor, Florida Institute of Technology**
- 1:30 pm Update about entomological research projects in the Navy Entomology Center of Excellence **Dr. Craig Stoops, Navy Entomology Center of Excellence, Naval Air Station, Jacksonville, FL**
- 1:50 pm Update about the DACS' mosquito control regulatory activities and NPDES **Mr. Steven Harrison, Entomology and Pest Control, DACS, Tallahassee, FL**
- 2:05 pm How to prevent bioterrorist's threats **Ms. Carla Lee, FBI Agent, FBI Biological Countermeasures, Jacksonville, FL**
- 2:20 pm House fly trapping: trap numbers and trap placement **Dr. Jerry Hogsette, Research Entomologist, USDA/ARS/CMAVE, Gainesville, FL**
- 2:35 pm Mosquitoes and sand fly research in Greece **Dr. Alex Chaskopoulou** (presented by Dr. Phil Koehler) **University of Florida, Dept. of Entomology and Nematology, Gainesville, FL**
- 2:50 pm Blood loss and public health consequences of bed bug infestations **Dr. Roberto Pereira, Research Scientist, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL**
- 3:10 pm Aquatic invertebrates and mosquito population surveillance in Guano River **Mr. John Henzler, Field Biologist, AMCD/DEP, St. Augustine, FL**
- 3:25 pm Break

Moderator: Mr. Bob Betts, Director, Escambia County Mosquito Control, FL

- 3:40 pm Overview of the American Mosquito Control Association **Mr. Doug Carlson, Past President, AMCA and Director, Indian River Mosquito Control District, Vero Beach, FL**
- 4:00 pm Several recent operational challenges at Indian River Mosquito Control District **Mr. Doug Carlson, Director, Indian River Mosquito Control District, Vero Beach, FL**
- 4:20 pm Lee County Mosquito Control Programs in 2011 **Mrs. Shelly Redovan, Deputy Director, Lee County Mosquito Control District, Ft. Myers, FL**
- 4:40 pm Environmental impacts of invasive plant species on mosquito habitats **Mr. Edward Northey and Mr. James McNelly, Director, Volusia Mosquito Control District, New Smyrna Beach, FL**
- 5:00 pm Mechanisms of mosquito larval and pupal mortality induced by Acoustic Larvicide® treatments and operational results from spring vernal pool

applications **Mr. Herbert Nyberg, New Mountain Innovations, 6 Hawthorne Road, Old Lyme, CT.**

5:20 pm End of session

Thursday, March 29, 2012

Animal Pest & Other Session:

Moderator: Dr. Phil Kaufman, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL

- 8:30 am Quick Bayt: Not so quick **Krista Seraydar and Phil Kaufman, Dept. of Entomology & Nematology, University of Florida, Gainesville, FL**
- 8:45 am Acaricide resistance in brown dog tick populations in Florida **Amanda Eiden, Phil Kaufman, and Faith Oi, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL**
- 9:00 am How do you measure dispersal in a pestiferous fly? **Dr. Phil Kaufman, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL**
- 9:15 am Repellent for livestock and poultry pests **Dr. Wes Watson, Professor, Dept. of Entomology, NC State University, Raleigh, NC**
- 9:30 am Update research on mosquito larval physiology and possible use for control **Dr. Paul Linser, Professor, University of Florida, Whitney Laboratory, St. Augustine, FL**
- 9:45 am Analysis of service requests at AMCD based on VCMS databases **Mr. Richard Weaver, Data Manager, AMCD, St. Augustine, FL.**
- 10:00 am Education and outreach progress at AMCD **Ms. Jessica Phillips, Education Specialist, AMCD, St. Augustine, FL**
- 10:15 am Mosquito population surveillance and control methods in Jiangsu, China **Dr. Hong-Liang Chu, Director, Department of Vector Control and Disinfection, Jiangsu CDC, Nanjing, China**
- 10:30 am Break

Technology Session:

Moderator: Dr. Paul Linser, Professor, UF/Whitney Laboratory, St. Augustine, FL

- 10:45 am Update MapVison software/database for mosquito control programs **Mr. Mike Reynolds, Leading Edge Association, LLC, NC**
- 11:00 am New technology and chemical update from ADAPCO **Mr. Derek Wright, ADAPCO, Sanford, FL**
- 11:15 am Overview of American LongRay equipment products **Mr. Brian Liang, American LongRay Company, San Francisco, CA**
- 11:25 am The LongRay fogger machine's testing report **Mr. Mike Smith, Biotech, AMCD, St. Augustine, FL**
- 11:35 am Overview of Clarke's new products for 2012 **Mr. Frank Clarke. Clarke Mosquito Control Products, Kissimmee, FL**
- 11:50 am Update about a new larval control technique for catch basins **Mrs. Candace Royals, Vector Sale Specialist, Valent BioSciences, Tampa, FL**

- 12:05 pm New products and formulation for 2012, All Pro Vector Group **Mr. David Sykes, Regional Representative, All Pro Vector**
- 12:10 pm Lunch break (provided by ADAPCO) and workshop end.
- 1:00 pm Tour of AMCD's facilities (upon request) **Dr. Whitney Qualls, Biologist, AMCD, St. Augustine, FL**

THE TENTH ARBOVIRUS SURVEILLANCE AND MOSQUITO CONTROL WORKSHOP

SPONSORED BY AMCD & USDA/CMAVE
ST. AUGUSTINE, FLORIDA, MARCH 26-28, 2013

(CELEBRATION OF TEN YEARS)

PROGRAM

Tuesday (March 26, 2013)

Panel Session:

Moderator: Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL

- 8:30 am Welcome & Introduction **Mrs. Jeanne Moeller, AMCD Commissioner Chairperson, Dr. Gary Clark, Research Leader, USDA/CMAVE, and Mr. Robert Betts, FMCA President**
- 8:40 am **Keynote Speaker:** Update of WNV and other mosquito-borne viruses in the USA **Dr. R. Nasci, Branch Chief, CDC, Ft. Collins, CO**
- 10:00 am The need for customized mosquito control: differences in genetics and behavior among US populations of *Aedes albopictus* **Dr. Dina Fonseca, Associate Professor, Center for Vector Biology, Rutgers University, NJ**
- 10:20 am Overview of the Medical Entomology Collaboration Program **Dr. Craig A. Stoops, Program Manager, Navy & Marine Corps Public Health Center, USDA/CMAVE, Gainesville, FL**
- 10:30 am The 3rd International Forum for Surveillance and Control of Mosquitoes and Mosquito-borne Diseases, Suzhou, China, May 27-31, 2013 **Dr. Rui-De Xue, Director, AMCD, St. Augustine, FL and Dr. Tongng-Yan Zhao, Professor and Director, Department of Vector Biology and Control, Beijing Institute of Microbiology & Epidemiology, Beijing, China**
- 10:40 am Break

Moderator: Dr. Dan Kline, Research Entomologist, USDA/CMAVE, Gainesville, FL

- 11:00 am Update of genotyping of *Aedes albopictus* species from Florida, compared to the strain from Malaysia **Dr. Azliyati Azizan, Assistant Professor, Dept. of Global Health, University of South Florida, Tampa, FL**
- 11:20 am The resurgence of *Aedes aegypti* in northeast Florida and potential implications **Lt. Jennifer Wright, Dr. Peter J. Obenauer, CDR, Entomologist, Navy Entomology Center for Excellence, Jacksonville, FL, Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL**
- 11:40 am USDA/Rutgers University area-wide management of Asian tiger mosquitoes **Dr. Gary Clark, Research Leader, USDA/CMAVE, Gainesville, FL**

- 11:50 am Overview of the Society of Vector Ecology & 6th International Congress of the SOVE **Dr. Douglas E. Norris, President-Elect -SOVE, Department of Molecular Microbiology and Immunology, The John Hopkins Malaria Research Institute, Bloomberg School of Public Health, Johns Hopkins University, Baltimore, MD**
- 12:00 pm Lunch break (provided by Clarke Mosquito Control)

Arbovirus & Malaria:

Moderator: Dr. Gary Clark, Research Leader, USDA/CMAVE, Gainesville, FL

- 1:00 pm Are arboviral epidemics puzzles or mysteries? **Dr. Jonathan F. Day, Professor, University of Florida, FMEL, Vero Beach, FL**
- 1:20 pm Arbovirus surveillance in Florida, 2012: an overview and the challenges of finding dengue cases in a year of West Nile **Stephanie M. Moody-Geissler, MPH, Arbovirus Surveillance Coordinator, Bureau of Epidemiologist, DOH, Tallahassee, FL**
- 1:40 pm WNV outbreak in Jacksonville, Florida, 2012 **Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL**
- 1:55 pm WNV outbreak and control response in Escambia County, FL **Mr. Robert Betts, Director, Escambia County Mosquito Control, FL**
- 2:10 pm Jacksonville Mosquito Control Response for WNV outbreak in 2012 **Mr. Richard Smith, Superintendent, Jacksonville Mosquito Control, FL**
- 2:25 pm Rapid screening of sero-conversion against WNV in sentinel chickens **Dr. Min-Lee Cheng & Dr. T. Steven Su, West Valley Mosquito and Vector Control District, Ontario, CA**
- 2:40 pm Dengue fever surveillance and control in Hong Kong, China **Mr. Ming-Ci Yuen, Pest Control Officer, Food & Environmental Hygiene Department, Hong Kong, China**
- 3:00 pm Break

Moderator: Dr. Uli. Bernier, Research Chemist, USDA/CMAVE, Gainesville, FL

- 3:20 pm Mosquito population surveillance and control methods in Jiangsu, China **Mr. Yufu Zhang, Department of Vector Control and Disinfection, Jiangsu CDC, Nanjing, China**
- 3:40 pm Accomplishment, challenge, and future of insecticide-treated bed nets against malaria in Zambia **Dr. Douglas Norris, Associate Professor, Department of Molecular Microbiology and Immunology, The John Hopkins Malaria Research Institute, Bloomberg School of Public Health, Johns Hopkins University, Baltimore, MD**
- 4:00 pm Integrated Vector Management for malaria control **Dr. Graham Mathews, Professor and Director, IPARC, Imperial College, London, UK**
- 4:20 pm Large scale control of *Anopheles crucians* with attractive toxic sugar bait barrier treatment and impact on non-target organisms in St. Augustine, Florida **W. A. Qualls, G.C. Muller, Edita E. Revay, V. Kravchenko, S. Efremova, A. Hausmann, M. L. Smith, J. Scott, J. Beier, and R-D. Xue**

- 4:35 pm Outdoor resting and sugar feeding behaviors of African malaria vectors
Dr. John Beier, Professor, University of Miami, Miami, FL
- 4:55 pm Update about permethrin-treated military uniform research **Dr. Uli Bernier, Research Chemist, USDA/CMAVE, Gainesville, FL**
- 5:15 pm Multi-scale modeling of malaria risk in northern South America **Ms. Temitope Alimi, University of Miami, Miami, FL** (presented by Dr. Whitney Qualls)
- 5:25 pm Update on traps for management of stable flies and house flies **Dr. Jerry Hogsette, Research Entomologist, USDA/ARS/CMAVE, Gainesville, FL**
- 5:45 pm End of session
- 6:00 pm Dinner and lecture (Holiday Inn, Beach Blvd, St. Augustine Beach)
Overview of DACS/DPI programs within the Bureau of Entomology, Nematology & Plant Pathology **Dr. Greg Hodges, Bureau Chief, Division of Plant Industry, Gainesville, FL**

Wednesday (March 27, 2013)

Biology, Attractants/Repellents:

Moderator: Dr. John Beier, Professor, University of Miami, Miami, FL

- 8:30 am Localization and expression of carbonic anhydrases in *Aedes aegypti* **Mr. D. Dixon and Dr. Paul Linser, Professor, University of Florida, Whitney Laboratory, St. Augustine, FL**
- 8:45 am Behavior modification of mosquitoes through CO₂ receptor agonists and antagonists **Dr. Michelle Brown, Research Scientist/Project Leader, Olfactor Laboratories, Riverside, CA**
- 8:55 am Development of field protocols for the evaluation of spatial repellents **Dr. Dan. Kline, Research Entomologist, USDA/CMAVE, Gainesville, FL**
- 9:15 am Toxicity of some essential oils to two species of adult mosquitoes **Dr. Joel Coats, Professor, Dept of Entomol, Iowa State University, Ames, IA**
- 9:35 am Survival of *Aedes albopictus* on native plants in Florida **Mrs. Alice Fulcher, Biologist, AMCD/DEP, St. Augustine, FL**
- 9:50 am Truck-mounted area application of pyriproxyfen targeting *Aedes albopictus* and *Aedes aegypti* in northeast Florida **Dr. Carl W. Doud, Technical Director, Navy Entomology Center of Excellence, Jacksonville, FL**
- 10:00 am Direct and indirect spray of the IGRs pyriproxyfen on emergence of *Aedes albopictus* in West St. Augustine, FL **Ms. Jodi Scott, Education Specialist, AMCD, St. Augustine, FL**
- 10:15 am Lethal ovitraps for *Aedes albopictus* control **Ephraim Ragasa and Phil Koehler, University of Florida, Dept. of Entomology and Nematology, Gainesville, FL**
- 10:30 am Break

Larval/Adult Control & Resistance:

Moderator: Dr. Douglas Norris, Associate Professor, Department of Molecular Microbiology and Immunology, The John Hopkins Malaria Research Institute, Bloomberg School of Public Health, Johns Hopkins University, Baltimore, MD

- 10:45 am Assessing efficacy of aerosol pesticide applications **Dr. Seth C. Britch, Dr. Kenneth Linthicum, and Mr. Robert L. Aldridge, USDA/CMAVE, Gainesville, FL**
- 11:00 am Aerial spray against outbreak of adult mosquitoes in St. Johns County, FL **Mrs. Kay Gaines, Base Station Supervisor, AMCD, St. Augustine, FL**
- 11:20 am Resistance and resistance management in biorational pesticides for mosquito control **Dr. T. Steven Su, Ecologist, Dr. Min-Lee Cheng, Manager, West Valley Mosquito and Vector Control District, Ontario, CA**
- 11:40 am Insecticide resistance monitoring in Polk County **Dr. Carl Bohene, Entomologist, Polk County Mosquito Control, FL**
- 11:50 am A new IGR mosquito larvicide - MOSQUIRON **Mr. Barry Tyler, Pestalto Environmental Health Services, Inc., Canada.**
- 12:10 pm Lunch break (provided by the Central Life Sciences)

Program, Public Relation, & Legislation:

Moderator: Dr. Dina Fonseca, Associate Professor, Center for Vector Biology, Rutgers University, NJ

- 1:10 pm DDT application and mosquito control **Dr. Gordon Patterson, Professor, Florida Institute of Technology**
- 1:30 am Public relations: Managing The Media **Mr. Joseph Conlon, Technical Advisor, AMCA**
- 1:50 pm Update about the Florida Mosquito Control Association **Mr. Robert Betts, President, FMCA and Director, Escambia County Mosquito Control, FL**
- 2:00 pm Update about the DACS' mosquito control regulatory activities **Mr. Michael Page, Chief, Bureau of Entomology and Pest Control, DACS, Tallahassee, FL**
- 2:15 pm FBI's WMD program overview **Mrs. B. Frost, Special Agent, Jacksonville, FL**
- 2:35 pm Do ground ULV sprays interrupt WN transmission cycle? **Dr. Peter Jiang, Medical Entomologist, Bureau of Pesticides, DACS, Tallahassee, FL**
- 2:50 pm Update on Greek mosquito and sand fly research projects **Dr. Alex Chaskopoulou, University of Florida, Dept. of Entomology and Nematology, Gainesville, FL** (presented by Dr. Phil Koehler)
- 3:10 pm Update about the American Mosquito Control Association **Dr. Roxanne Connelly, President-AMCA, Associate Professor, University of Florida, IFAS/FMEL, Vero Beach, FL**
- 3:25 pm Break

Moderator: Mr. Bob Betts, Director, Escambia County Mosquito Control, FL

- 3:40 pm Modeling a potential mechanism for RFV outbreak in Egypt: implication for control **Mr. Harold Gill, University of Miami, Miami, FL**
- 3:50 pm Requesting FEMA reimbursement after a disaster declaration **Ms. Marah Clark, Entomologist, Jacksonville Mosquito Control, FL**

- 4:00 pm Manatee County Mosquito Control District program **Mr. Mark Latham, Director, Manatee County Mosquito Control District, FL**
- 4:20 pm Volusia Mosquito Control Programs **Mr. James McNelly, Director, Volusia Mosquito Control, Daytona Beach, FL**
- 4:40 pm Beach Mosquito Control Program **Mr. Dale Martin, Entomologist and Mr. James Clauson, Director, Beach Mosquito Control District, Panama City, FL**
- 5:05 pm Polk County Mosquito Control Pprogram **Mr. Michael Mahler, Director, Polk County Mosquito Control, FL**
- 5:20 pm Florida Keys Mosquito Control Program **Michael Doyle, Director, Florida Keys Mosquito Control District, Key West, FL**
- 5:40 pm Mosquito control program in Ecuador **Ms. D. Naranjo, University of Miami, Miami, FL**
- 6:00 pm WNV control in Texas, 2012 **Mr. Frank Clarke, Vice President, Clarke, Kissimmee, FL**
- 6:20 pm End of session

Thursday (March 28, 2013)

Other Pests, Vectors, & Methods:

Moderator: Dr. Phil Koehler, Professor, University of Florida, Dept. of Entomology and Nematology, Gainesville, FL

- 8:30 am Medical and veterinary significance of bed bugs **Dr. Roberto Pereira, Research Scientist, Presented by Dr. Phil Koehler, Professor, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL**
- 8:50 pm Bedbugs and their movement on surfaces **Ben Hottel and Phil Koehler, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL**
- 9:05 am Evaluation of resistance mechanisms in brown dog tick (*Rhipicephalus sanguineus*) populations in Florida **Amanda Eiden, Michael Dark, Robert Miller, Faith Oi, and Phil Kaufman, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL**
- 9:20 am Picking up the scent of a new method for brown dog tick management **Lucas Carnohan, Emma Weeks, Sandra Allan, and Phil Kaufman, Dept. of Entomology and Nematology, University of Florida, Gainesville, FL**
- 9:35 am Laboratory and field evaluation of tick repellents **Jodi Scott. Rui-De Xue, AMCD, St. Augustine, FL**
- 9:50 am Analysis of pesticide application at AMCD based on VCMS databases **Mr. Richard Weaver, Data Manager, AMCD, St. Augustine, FL.**
- 10:05 am Update about the ATSB **Mrs. Laura Uggerholt, CEO & Partner, Universal Pest Solution, Dallas, Tx**
- 10:25 am Break

Technology& New Products:

Moderator: Dr. Paul Linser, Professor, UF/Whitney Laboratory, St. Augustine, FL

- 10:40 am Update about the American LongRay equipment products **Mr. Brian Liang, American LongRay Company, San Francisco, CA**
- 11:00 am The LongRay truck-mounted thermal fogger machine's testing report **Mr. Mike Smith, Biotech, AMCD, St. Augustine, FL.**
- 11:20 am Overview of Clarke's new products for 2013 **Mr. Frank Clarke, Vice President, Clarke, Kissimmee, FL**
- 11:40 am Overview of the Central Life Sciences' products for 2013 **Mr. Larry Smith, Central Life Sciences, Mount Laurel, NJ**
- 11:55 am Overview of UNIVAR products for 2013 **Mr. Jason E. Conrad, Industry Specialist, UNIVAR, GA**
- 12:05 pm Lunch break (provided by ADAPCO) and workshop end.
- 1:30 pm Tour of AMCD's facilities (upon request) **Mrs. Alice Fulcher, Biologist, AMCD, St. Augustine, FL 500 e, Fl. 32080 (904) 471-3107**

