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# ABSTRACTS

of Submitted Papers, Posters, and Symposia Presentations



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# The 2008 AMCA Memorial Lecture Honoree: Dr. Andrew Spielman, 1930-2006

Dr. Andrew (Andy) Spielman died unexpectedly on December 20, 2006, after a brief illness that began while he was attending the President's Malaria Summit in Washington, D.C. His passing marked the end of a highly distinguished 48- year teaching and research career at the Harvard School of Public Health in Boston. Andy was one of the most productive, published and innovative medical entomologists of all time. After completing his PhD on *Culex* autogeny with famed medical entomologist Lloyd Roseboom at Johns Hopkins, Andy entered the

Navy and was stationed at Guantanamo, Cuba working on eye gnat and other pest control problems. This practical work along with a brief stint at TVA's Wilson Dam in Alabama studying the disappearance of malaria had a lasting impact on Andy's approach to research. He took an unusually holistic view and always had one foot in the field and the other in the laboratory as he searched for the useful application of his research findings. Andy began his Harvard career working on mosquitoes and with Carroll Williams and John Law (creators of the first juvenile hormone

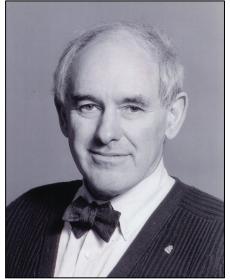
analog) he conducted the first demonstration of the ability of JH mimics to control mosquito larvae through arrested development. This 1966 paper was the first of 7 papers published in *Science* and one of over 360 others. During the initial phase of his career from 1959 to 1975, Andy's seminal research focused primarily on *Culex pipiens* and *Aedes aegypti* physiology, population biology, and reproduction along with mosquito arbovirus interaction. 1975 marked the beginning of the second phase of Andy's career when he published his first paper on tick-borne babesiosis. Although some work on mosquitoes and

filaria continued, the main focus of Andy's lab became ticks and tick-borne infections. He is most recognized for his pioneering work in unraveling the dynamic interactions between *Ixodes* ticks and the organisms causing babesiosis and Lyme disease. On these subjects, he became the world's leading expert. In the 1990's he began migrating back to his mosquito roots and the final phase of his career. His interests in malaria resurfaced and his most novel research on Eastern equine encephalitis began. After West Nile appeared, Andy dusted off his early papers on *Culex* 

pipiens and applied them to the puzzles of this new, emerging disease. Andy traveled and collaborated with scientists all over the world, and leaving left his mark and unique perspectives wherever he went. Many of his ideas invoked controversy and Andy relished the lively debates they precipitated. He made people think, and think deeply, about vector-borne diseases and how best to control them in a sustainable way. Unfortunately, the final controversy on the role of maize pollen in malaria dynamics in Africa will have to be resolved by others. Andy carefully molded and mentored

an entire generation of outstanding medical entomologists who today occupy important

positions throughout the world. Andy received many awards, the most notable being the AMCA's Medal of Honor, a 10-year career award from NIH, election as president of the Society of Vector Ecology and the ASTMH's coveted Hoogstraal medal. His co-authored 2001 book "Mosquito: a natural history of our most deadly foe" was so popular it has been translated into many languages. It explodes with Andy's unique and enthusiastic views on mosquitoes and the related stories that only Andy can tell.



Dr. Andrew Spielman

# The 2008 AMCA Memorial Lecturer: John Edman



Dr. John Edman

Dr. John Edman retired in late 2004 after a 40-year academic career of teaching, research, and administration at the University of California at Davis (1999-2004), the University of Massachusetts at Amherst (1975-99) and the Florida Medical Entomology Laboratory at Vero Beach (1964-75). He chaired the Entomology Department at the University of Massachusetts for 7 years and was Director of the Center for Vectorborne Diseases at UC Davis for 5 years. A former AMCA president and recipient of the Medal of Honor, he has been a member of the AMCA since 1959. He is also a past president of the Society for Vector Ecology and a past Chair of Section D and member of the Governing Board of the Entomological Society of America. He received the L.O. Howard award and the Outstanding Teacher Award from the Eastern Branch of the ESA. In 2004, he was awarded the prestigious Hoogstraal Medal given by the American Society of Tropical Medicine and Hygiene. Dr. Edman trained 25 doctoral and masters students while at UMASS; his first doctoral student (Roger Nasci) is also a former AMCA president. In addition to a wide array of research interests and nearly 200 research publications, reviews and book chapters, Dr. Edman, along with Dr. Bruce Eldridge, co-authored the widely used text for graduate and advance undergraduate courses in medical and veterinary entomology entitled: Medical Entomology - A Textbook on Public Health and Veterinary Problems

Caused by Arthropods. Many of his publications focused on the ecology and behavior of mosquito vectors and transmission of malaria, filaria, and arboviruses including dengue, West Nile, St. Louis encephalitis, and eastern equine encephalitis. He is perhaps most recognized for his innovative research on the defensive (anti-mosquito) behavior of vertebrate hosts; interrupted and multiple bloodfeeding behavior; and demonstration that certain Culex vectors shift their feeding patterns from avian amplification hosts to significant feeding on mammals during the latter part of the arbovirus transmission season when human cases occur.



Andy Spielman and John Edman in the field.

# **Oral Presentation Abstracts**

### 1 Introduction to the Operational Side of Mosquito Control – The Application of IPM Techniques for Effective Mosquito Control Symposium

Bill Reinert, reinert\_william@aclink.org

Presenters will discuss various aspects of integrated pest management practices involved in quality mosquito control from an operational level. Topics discussed will include surveillance, larval and adult control materials and methods, water management, biological control, and community outreach and education.

## 2 Aedes albopictus, one year's experience - twenty one times

Matthew Yates, myates@brgov.com

Aedes albopictus was first discovered in East Baton Rouge Parish in 1986 at a tire recycling facility. It rapidly spread throughout the parish and has become the most serious pest mosquito in urban areas of the parish. Mosquito control personnel have isolated EEE virus and West Nile virus from Ae. albopictus on several occasions, so it is also a potential disease vector in the parish. Mosquito control personnel have implemented a variety of physical control methods and employed several public education techniques over the last 21 yr in an effort to control this mosquito. Most of the efforts failed or only had limited success. This presentation will highlight 21 yr of disease surveillance, control efforts, and public education techniques aimed at Ae. albopictus in East Baton Rouge Parish, Louisiana and review what has worked and what has not.

#### 3 Pre-flood treatments against floodwater mosquitoes

Tom Floore, tom.floore@mosquito.org

Pre-flood applications of methoprene and other larvicides have been used in the control of floodwater mosquitoes for over 40 yr. In this presentation, we will define pre-flood or pre-hatch applications, look at some early studies first using this method, and review some PHEREC studies. PHEREC has been involved in pre-flood methoprene studies for over 30 yr.

### 4 Larviciding in MI: The silent tool of IPM in mosquito control

Randall Knepper, Randy@scmac.org

Saginaw County, MI, has a unique blend of urban and rural larval habitats, resulting in innovative larviciding methods. Larviciding programs discussed will include aerial application for spring *Aedes* mosquitoes; treatment of *Culex* mosquitoes in urban catch basins with customized mopeds; cooperative relationship with local health department in controlling mosquito larvae in abandoned swimming pools; larval control of various container species in urban neighborhoods; and efforts to control floodwater mosquitoes.

#### 5 Tree-hole treatments to reduce dog heartworm incidence in Salt Lake City

Sammie Dickson, sdickson@slc-mosquito.com

Ochlerotatus sierrensis, the western tree-hole mosquito, was first identified in Salt Lake City in 1987. While the finding of a new species in the city was a curiosity, it turned out not to be a coincidence that the first case of Dirofilaria immitis occurred that same year. Ochlerotatus sierrensis spread quickly throughout Salt Lake City in the following years until, in 1992, the incidence of dog heartworm and service requests for small biting mosquitoes in the city reached levels that required the Salt Lake City Mosquito Abatement District to begin a new program. The first attempt at controlling Oc. sierrensis began with filling holes and has since evolved into a yearly treatment program involving the placement of a timed-release briquette in more than 2,700 tree-holes.

# The production and distribution of mosquitofish and guppies at the Sacramento-Yolo MVCD Demetri Dokos, ddokos@FIGHTtheBITE.net

Techniques are provided for the production and distribution of mosquitofish and guppies at the Sacramento-Yolo Mosquito & Vector Control District. Pond design, algae bloom, over-wintering, culturing, and stocking fish for mosquito control purposes are discussed.

#### 7 Surveillance traps in NJ mosquito control programs

Ary Farajollahi, afarajollahi@mercercounty.org, and Greg Williams

A variety of surveillance tools is utilized in NJ to monitor adult mosquito population densities, species compositions, and field infections for arboviruses. Efficiently monitoring a vector of choice requires the use of the appropriate tool. Surveillance tools generally include resting boxes, lights traps, carbon dioxide-baited traps, and gravid traps. In response to rising complaints from high levels of *Aedes albopictus* mosquitoes, BG Sentinel traps were also evaluated. We discuss the use of surveillance traps in NJ county mosquito control programs, and how they may be utilized to make informed decisions on controlling nuisance and disease carrying mosquitoes.

### 8 Pesticide use protocols in Suffolk County, NY

Dominick Ninivaggi, dominick.ninivaggi@co.suffolk.ny.us

As part of its Vector Control and Wetlands Management Long Term Plan, Suffolk County has developed protocols to describe under what conditions pesticides will be used. For adulticides, surveillance thresholds and methods have been established that could trigger treatments, as well as countervailing factors that would lead to a "no treatment" decision. A similar approach was taken with larvicides with the addition of criteria for choice of larvicide. The larvicide guidelines include a preference for bacterial products when these are likely to be effective, while preserving the option of methoprene products when necessary. The protocols were developed to meet demands for objective criteria for the use and choice of pesticides and for increased program transparency. Selecting the appropriate threshold level for treatment continues to be controversial because of difficulties in defining what population levels of a public health pest are acceptable.

### 9 Introduction to Legislative & Regulatory Symposium

William Meredith, William.Meredith@state.de.us

Through a group of speakers and a question and answer session, this symposium will provide the opportunity for AMCA members to learn the current status of federal legislative and regulatory issues of importance to mosquito control.

### 10 WNV/Epidemiology & Laboratory Capacity (ELC) grant funding issues

Tracy Badsgard, tab7@cdc.gov

### 11 Will you be sued under the Endangered Species Act or Clean Water Act in 2008?

Karl Malamud-Roam, kmr@ccmvcd.net

## 12 USFWS refuge mosquito issues & stormwater management

William Meredith, William.Meredith@state.de.us

### 13 Legislative and regulatory issues from a Washington perspective

Ed Ruckert, eruckert@mwe.com

# 14 Comparative capture rates of adult mosquitoes by light trap and human landing collection methods Donald Barnard, don.barnard@ars.usda.gov, and Gregory Knue

The landing rates (LR) of Anopheles quadrimaculatus, Culex nigripalpus, Cx. quinquefasciatus, Ochlerotatus triseriatus and Aedes albopictus on human hosts were compared with capture rates of the same species by CDC light traps augmented with CO<sub>2</sub>. Significant associations with LR were observed among the day-to-day responses to CDC by An. quadrimaculatus and Cx. quinquefasciatus and among the hour-by-hour (over 24 h) responses to same by all species except Oc. triseriatus. CDC traps typically underestimate LR, depending on the mosquito species and time of day, but improved precision in these estimates is achieved by the identification/removal of outlier responses and the fit of transformed data for each species to polynomial models.

# You've come a long way Aedes: A fifty-year review of surveillance methods and mosquito fauna in MN Diann Crane, dianncrane@mmcd.org, Sandy Brogren, and Carey LaMere

The Metropolitan Mosquito Control District has proudly served its MN citizens for 50 yr. Since the District's inception in 1958, there have been many improvements, especially in the methods of mosquito surveillance. In the beginning, we used primarily New Jersey light traps and dippers. Over the years, we added a variety of methods to answer specific questions regarding abundance, annoyance, disease activity, and locations for mosquito control operations. This paper will discuss the evolution of surveillance methods and the changes in mosquito fauna in 50 yr at MMCD, including the first occurrence of *Aedes japonicus* in MN.

# 16 Seasonal distribution of Aedes japonicus in NJLT collections

Tom R. Wilmot, twilmot@co.midland.mi.us

Aedes japonicus was first seen in Midland County, MI, in 2004. It is now well established in the county and is among the most commonly identified mosquitoes from container-habitat samples. Over the last few years, we have seen a distinct trend in NJLT collections of Ae. japonicus; very few are collected during the spring or summer with a significant increase in numbers collected in September to October. Further studies are planned to determine if this trend correlates with actual numbers of mosquitoes present or reflects a relative change over the season in response of this species to light traps.

# 17 A study on the eggs of Aedes aegypti and Aedes albopictus in TX

Catherine Z. Dickerson, catherine.dickerson@ars.usda.gov, and Jimmy K. Olson

Causative influences that impact the separation of *Aedes aegypti* and *Ae. albopictus* populations in different geographic areas were determined. The eggs of *Ae. albopictus* and *Ae. aegypti* collected from McAllen and Brownsville, TX, and laboratory populations of these 2 species were subjected to 25 temperature and relative humidity combinations for up to 3 mo. In most treatments, *Ae. aegypti* eggs had a greater percent hatch than *Ae.* 

albopictus. The surface area, volume, and surface-area-to-volume ratio of Ae. aegypti and Ae. albopictus eggs with and without the chorionic egg pad, and the size of the chorionic egg pad were calculated for 50 eggs of each species of mosquito from the afore-mentioned populations. Aedes aegypti had a larger egg volume, and a larger surface area, but it is likely their larger chorionic egg pad compensates for this high surface-area-to-volume ratio by holding moisture along the egg's surface and that the egg pad is associated with the high desiccation resistance seen in Ae. aegypti eggs.

# 18 Aedes albopictus male longevity, dispersal and mating capacity: Good enough for cost/benefit site application?

R. Bellini, rbellini@caa.it, M.Calvitti, and S.Urbanelli

The spread of *Aedes albopictus* (Skuse) is continuing in Europe, Africa, and America continents. Recently an epidemic of chikungunya virus vectored by *Ae. albopictus* took place in northern Italy with more than 200 human cases. Control of this species with conventional methods appears particularly difficult mainly because of the large amount of breeding sites available in private areas. Until now, community participation has shown unsatisfactory results. We considered this species a suitable target for Sterile Insect Technique application because of its recent introduction, the urban-related distribution, and the low active dispersal potential. A small pilot rearing facility targeted to produce up to 100,000 male pupae weekly has been established. The sterilization is performed on the pupal stage with gamma irradiation. Studies on males' longevity and mating capacity have been carried out in the laboratory (cages 40x40x40 cm). Sterile males' competitiveness and longevity have been investigated in field conditions greenhouses (5x8x2.8 m) and males' longevity and dispersal have been evaluated in urban areas by mark-recapture technique using 2 parallel methods: 1. releasing reared pigment-marked adults, and 2. releasing pupae of a *Wolbachia* removed strain (aposymbiotic).

19 Seasonal distribution and species composition of daytime biting mosquitoes in different ecological zones Waseem Akram, areeba14@yahoo.com, Faisal Hafeez, Unsar Naeem Ullah, Aftab Hussain, and Jong-Jin Lee

Severe summer temperatures of 50°C with average fluctuating to around 40-44 °C is considered one of the factors in increasing the distribution habitats of mosquitoes. Major mosquito activity in various ecological zones of Pakistan starts from February and continues until mid-December, Mosquitoes, particularly the daytime biting Aedes, constitute a healthy fauna that is distributed in different ecological areas. The populations of Aedes peak between May to September, while those of *Culex* and *Anopheles* peak from March to September. Average population counts for Anopheles, Culex, and Aedes show a ratio to 50:90:18 from May until November. A population fluctuation reduces activity at 45-50 °C with RH 45%. For most of the mosquito species, more individuals are collected during the "high season" months (May to September) than in the "low season" months (January to mid-February). In ecological zones with summer temperature of 44±5 °C during June to July mosquito activity is suspended at RH 70%. The populations of Culex (36.8%), Anopheles (32.09%), and Aedes (26.3%) begin to appear in greater numbers in the month of August with temperature more or less in the range of 40±2 °C. Population estimates (through standard prototype CDC and Bio sentinel traps) and species composition of Aedes in forests indicate a rapid increase in the population of Aedes albopictus (52.3%), Ae. aegypti (19.1%), along with Ae vittatus (28.5%) followed by rain in July. Areas positive for Ae. albopictus are identical to the population levels (1:1) and distribution ranges of Ae vittatus. However, Ae, aegypti are totally isolated from all Ae albopictus positive areas, visible from August to September in limited collection sites. The population levels, seasonal distribution, habitats, and areas of adult activity have been marked on a Global Positioning System used for reference and species composition of Anopheles, Culex, and Aedes as they are temperature and RH dependent. The expansion of the active period has come about from the global meteorological changes. Therefore, the possibilities of mosquitoes being disease vectors has ultimately risen, resulting in the spread of emerging diseases.

# An evaluation of the efficiency of ground and elevated canopy trapping to estimate *Culiseta melanura* population in Hillsborough County, FL, USA

Carlos A. Fernandes, fernandesc@hillsboroughcounty.org, and Donald Hayes

We tested the effectiveness of  $CO_2$ -baited Center for Disease Control and Prevention light traps elevated in the tree canopy (5.2-6.0 m) compared with light traps placed at ground level (1.5 m) for collecting *Culiseta melanura* as an indicator of eastern equine encephalitis virus (EEE) activity during 2006 and 2007. Collection was part of a surveillance program that encompasses 15 different sites (2 traps per site) preferably located along the edges of permanent freshwater swamps. The major goals of the project were to identify a trap or trapping method to consistently monitor the presence of Cs. melanura and to develop a pulley system to elevate and lower the test traps at the different locations. Other genera of mosquitoes reported were Culex, Anopheles, Aedes/Ochlerotatus, and Coquillettidia. Monthly data for the study period from each collection site will be presented, along with some of the environmental factors affecting the operation (i.e., rainfall, wind speed).

# 21 Comparative population genetics: *Culex restuans* vs. *Cx. pipiens* in the eastern US Dina M. Fonseca, dinafons@rci.rutgers.edu, and Laura D. Kramer

Culex restuans is an abundant local species that appears to occur seamlessly from north to south in the eastern US. This species starts its activity earlier in the year than Cx. pipiens (May/June vs. July), so it has been proposed that it is an important vector of WNV early in the transmission season. Although during the mosquito season Cx. restuans

are often collected together with *Cx. pipiens* in urban areas in New York State often making their relative importance as vectors difficult to resolve; during the winter, diapausing populations found in human associated structures like sewers and basements are uniquely comprised of *Cx. pipiens*. In contrast, we do not know where *Cx. restuans* survive the winter. One possibility is that *Cx. restuans* overwinters in rural areas (possibly in the leaf litter) and reinvades cities every spring. We have made a comparison of genetic diversity of *Cx. pipiens* and *Cx. restuans*. We currently have 19 molecular markers (microsatellites) optimized for *Cx. restuans*. Preliminary analyses indicate the absence of winter driven bottlenecks in *Cx. restuans*, which supports the hypothesis that this species diapauses in very large numbers possibly in a myriad of small isolated natural cavities.

### 22 Predatory and parasitic mites associated with mosquitoes in NJ

Anwar L. Bilgrami, anwarbil@rci.rutgers.edu, Gregory M. Williams, and Peter Bosak

A central premise of biological mosquito control is that the density of mosquitoes is reduced to stable equilibrium levels by using biological control agents. Parasitic and predaceous mites, of which several species are aquatic in nature, may possess potential to control mosquito populations. To determine mite prevalence and their association with mosquitoes in the state of New Jersey, over 1,000 adult mosquitoes infested with mites were isolated from light and gravid traps during 2007. Mites belonging to 5 subfamilies (i.e., Histiostomatinae, Rhizoglyphinae, Arrenurinae, Thyadinae and Anystinae) were identified. Three [i.e., Arrenurus (Arrenurinae), Thyas (Thyadinae), and Anystis (Anystinae)] were identified to generic level. Mosquito genera, Culex, Culiseta, Aedes, and Coquillettidia, were found infested with these mites. They were most common to Coquillettidia perturbans followed by Aedes and Ochlerotatus spp. Most mites were attached between head and thorax or on the upper abdomen. Presumably, they attached to an emerging mosquito from pupae at the water surface. The infestation rates ranged between 1 and 30 mites per mosquito. Histiostomatid mites are filter feeders, requiring an aqueous environment to survive. Their association with mosquitoes is therefore not too surprising. Anystis sp. is more or less terrestrial, being found in leaf litter. It is an aggressive and active predator of anything it can overcome. The present study suggests parasitic and predaceous mites are prevalent in New Jersey and may be exploited to manage mosquito populations.

# Surveillance, prevention, and control of mosquitoes and mosquito-borne diseases in Cape May County, NJ Anwar Bilgrami, anwarbil@rci.rutgers.edu, and Peter Bosak

Mosquitoes threaten public health and create social and economic problems in our communities. Prevention and control of mosquitoes and mosquito-borne diseases are accomplished through a comprehensive integrated mosquito management (IMM) approach. Integrated mosquito management employs techniques targeted against mosquitoes at their most vulnerable stages. Mosquito surveillance entails an assessment of species and populations as well as habitats that produce and harbor them. Surveillance is an essential component of IMM in the control and prevention of mosquitoes and mosquito-borne diseases. Birds and mammals can be important sentinels for mosquito-borne viruses which may provide early warning to prevent human cases. Larval control involves a variety of techniques that include water management and biological (parasites, predators, and pathogens) and chemical (physical, organic, and inorganic) control. Water management exploits the fact that larvae require water to complete a part of their life cycle. Removal of this habitat simply translates to no mosquito production. Biological control uses mosquito predators, parasites, and pathogens in the aquatic environment when removal of standing water is not an option. Options for adult control are far more limited and include trapping and chemical spray applications. Integrated mosquito management recognizes that no one-control strategy is the best for all situations, and that all have their advantages and drawbacks. Integrated mosquito management allows the user to pick and choose the set of strategies that best works for their particular situation.

### 24 Adulticiding activities in Harris County, TX (2007)

Kyle Flatt, kflatt@hcphes.org, James Self, Salvador Rico, Pamela Stark, and Rudy Bueno, Jr.

Harris County currently utilizes multiple adulticiding methods in its extensive efforts to control mosquitoes during disease outbreaks. The establishment of West Nile virus (WNV) in 2002 has greatly contributed to the evolution of these strategies that are primarily directed against *Culex quinquefasciatus* Say, the principal vector of WNV and Saint Louis encephalitis in the region. The purpose of this presentation is to describe the methods used by the Division and the rationale behind their usage.

# 25 ULV applications via all terrain vehicles in Morris County, NJ

Mark Vlazny, mvlazny@co.morris.nj.us, Marc Slaff, Kris McMorland, and Jeff Donnelly

Adult mosquito control in Morris County had been limited almost exclusively to truck-mounted ULV operations. This meant that adult mosquitoes would reach extremely high levels in residential areas before control was possible. Several years ago, we began testing all terrain vehicles with small ULV machines to control floodwater species as they emerged from their larval habitats with excellent results. In addition to controlling mosquitoes before they enter neighborhoods, the already low level of possible pesticide exposure was reduced even further.

# 26 Aerial adulticiding: Aspects to consider for the promotion of successful operations Mark Latham, manateemcd@aol.com

Aerial adulticiding for the control of adult mosquitoes has been conducted successfully for over 50 yr. However, there is always room for improvement, particularly in these times of heightened public awareness with the advent of increased mosquito control operations in response to West Nile virus and the resultant scrutiny over increased pesticide usage. The success of operations relies not only on understanding the correct methodology to target the species of concern, but also on evaluating each and every application to better understand why successes and failures occur. Mosquito control is also very reliant on developing the public's trust in understanding both the need for and the safety of the proposed operations. This presentation will touch upon aspects of aerial adulticiding that must be considered, including: Equipment, timing, flight parameters, adjustments for different target species, efficacy evaluation, public notification, new technologies, and alternative strategies.

# 27 The performance of AquaHalt in the operational program of Merced County MAD Allan Inman, mcmadmanager@vtlnet.com, Rod Gomes, Bill Jany, and Ben Goudie

Adulticiding is a key component of the West Nile virus prevention strategy of the Merced County Mosquito Abatement District. Early, late, and season-long adulticiding targeting *Culex tarsalis* populations resulted in 80%, 83%, and 53% reductions from the 5-yr historical average over 6-wk peak populations periods (wk 39-44) in the fall of 2004, 2005, and 2006, respectively. Merced County reported the lowest WNV infection rates in *Culex* mosquitoes for 2005 and 2006 in the Central Valley of CA. The District has tested various "Aqua" adulticiding products since 1997. AquaHalt (5% pyrethrins, 25% PBO) was operationally evaluated in 2007. Ground adulticiding cage trials produced 12-h mortalities of 100% in 5 of 6 replications over a 2-night period. Aerial treatments recorded 12-h mortality of 88% - 98%. AquaHalt has been added to the line of pyrethrin adulticides routinely used by the Merced County Mosquito Abatement District.

# Mosquito reduction best management practices: Implementation, evaluation, and community outreach Joel Buettner, jbuettner@FightTheBite.net

Mosquito source reduction is widely accepted as an important and effective component of an integrated mosquito management program yet can be difficult to achieve on a large scale. Building upon a long history of source reduction, water management, and community outreach efforts, the Sacramento-Yolo Mosquito and Vector Control District's Ecological Management Department has developed a process to encourage the widespread implementation of mosquito reduction best management practices (BMP) by identifying significant mosquito sources, working cooperatively with land owners, providing assistance in implementing BMP, and utilizing enforcement actions under the CA Health and Safety Code when necessary. Following an extensive public review process, the mosquito reduction BMP program was approved by the District in April 2007 and has successfully addressed nearly 80 significant mosquito sources in its first season. This program has helped to identify and give rise to several specific BMP implementation efforts such as a fall flooding program that promotes later fall flooding of seasonal wetlands and duck hunting clubs, a cemetery vase program that promotes the use of super-absorbent polyacrylamide in flower vases, a beaver dam program that addresses flooding caused by beaver dams, and a proposed grant program to provide assistance for BMP implementation projects.

# 29 Climate changes (not weather), and adaptations of Northeast Massachusetts Mosquito Control's Wetlands Management Program

Emily Sullivan, nemmc.edws@comcast.net

The Northeast Massachusetts Mosquito Control and Wetlands Management District is experiencing tremendous pressure to keep up with rapidly changing climates in the Northeast. The term "climate" is used loosely here and refers to external dynamics which have and continue to define this District's operations. Everything from the high-speed pace of computer technology, a hot political atmosphere, a tightening regulatory environment, the introduction of vector species and increased disease, and the need to show accountability as never before has forced this District to come up with new approaches and consider alternate strategies. This presentation will cover the ongoing evolution of the NEMMCWMD's wetland management program with a brief overview from the early 60's to present day. Follow the ongoing saga of open marsh water management (OMWM), from its debut in Essex County, MA, in the mid 1980's to its recent and hopefully temporary fall in 2006 -2007. Other subjects covered include a different look at grid ditching, interagency efforts to develop fresh water BMPs, potential restoration partnerships, historic bids for water quality permits, endangered species issues, and more. Highlights include a showcase of fun and exciting LGP equipment for the field worker in all of us.

# 30 Source reduction along FL's Indian River Lagoon: Controlling salt-marsh mosquitoes while enhancing the resource

Douglas Carlson, dcarlson1@hotmail.com

Over the past 80+ yr along east-central FL's brackish Indian River Lagoon, source reduction (=permanent control) has played a vital role in IPM programs aimed at controlling the vast populations of *Aedes taeniorhynchus* and *Ae. sollicitans*. From ditching, to dredging and filling, to the creation of impoundments, this work has been effective yet at times controversial. This presentation will review the scientific and political solutions to controlling these

mosquitoes with a minimum of pesticide use. Examples of recent challenges in accomplishing these tasks will be discussed.

# 31 Introduction to State Regulatory Issues Symposium

Kevin Sweeney, sweeney, kevin@epa.gov, and Joseph Conlon

This session will be a forum for discussion of regulatory issues of interest to the AMCA membership that will be facilitated by the AMCA Technical Director, Joseph Conlon. The panel will consist of state and federal regulators, an industry representative, AMCA representative, and a CDC scientist. The panel will address a series of questions in the areas of pesticide safety; interpretation, usefulness, and enforcement of pesticide product labeling; environmental fate or effects issues together with unscripted questions form the audience as they relate to mosquito/vector control programs and regulatory activities.

### 32 Issues of regulatory importance

Steve Blunt, CO Dept. of Agriculture
James Clauson, FL Dept of Agriculture & Consumer Services
Charles Moses, NV Dept. of Agriculture
Bob Hays, ID Dept. of Agriculture
David Brown, Sacramento-Yolo Mosq. & Vector Control District
Representative, EPA Region 9
Maria, Herrero, Valent BioSciences Incorporated
Janet McAllister, CDC

## 33 Facilitated panel and audience discussion

Joseph Conlon, AMCA Technical Advisor

# 34 Culex pipiens hybridization, feeding behavior, and parity rates on late season West Nile virus activity in DE Linda-Lou O'Connor, lindalou@udel.edu, John B. Gingrich, Dina Fonseca, and Thomas R. Unnasch

We examined various factors in *Culex pipiens* that influenced enzootic WNV activity in DE. Collections of mosquitoes were made at 6 locations that previously showed elevated epizootic activity in 2003 and 2004. We performed longitudinal comparisons of these 6 sites in 2006, 3 of which showed continued enzootic WNV activity based on virus-positive mosquitoes and sentinel chicken antibody seroconversions. The *Cx. pipiens* populations sampled from three sites were analyzed using 8 microsatellite DNA markers to determine the extent of hybridization between *Cx. pipiens* form *pipiens* and *Cx. pipiens* form *molestus*. Blood-meal preference for *Cx. pipiens* hybrids was carried out using mosquito choice tests in the laboratory and by using a PCR-heteroduplex assay protocol for blooded females collected in the field. Parity rates over the mosquito season (June to October) were also examined to determine the age structure and daily mortality rate of the population. Parous females increased from 52% in June to 98.6% (n=30) in late September indicating an older population later in the season. By late August, daily mortality was less than 10% and remained low through the end of the biting season. Other comparisons included mosquito species composition and abundance at all 6 sites.

# 35 Integrating operational data to characterize mosquito control operational areas for disease management in Harris County, TX

Rudy Bueno, Jr., rbueno@ hcphes.org, Kyle L. Flatt, and Salvador Rico

Harris County, TX is the 3<sup>rd</sup> largest county in the US and includes Houston, the 4<sup>th</sup> largest city. The total area is 1,788 square miles and is very diverse. The Mosquito Control Division, a part of the Harris County Public Health & Environmental Services Department, is responsible for countywide mosquito control with the primary focus being surveillance and control of mosquito-borne diseases such as SLE and WNV. Due to the diversity of the landscape features, environmental parameters, demographics, and the presence of mosquito-borne diseases both currently and historically, the MCD decided to launch an area characterization project to help provide a preliminary disease risk assessment of selected mosquito control operational areas (MCOA). Current and historical data will be used for this assessment. To date, several MCOA's have been completed with several others initiated. The purpose of this presentation is to highlight the importance of incorporating operational data for the initiation and development of this project. This will enhance prevention and control efforts of mosquito-borne diseases to protect the public health of the residents of Harris County.

#### 36 Successful suppression of West Nile virus activity

Henry Lewandowski, hblewandowski@chathamcounty.org, Robert Moulis, Rosmarie Kelly, E. Scott Yackel, Mark Hansen, Daniel G. Mead, Jeffrey Heusel, and Jennifer Russell

A West Nile fever/encephalitis epidemic in 2003 demonstrated what aspects of a local mosquito surveillance and control program were effective in monitoring and limiting human disease. Program improvements, implemented from 2004-2007 were tested for the first time during the 2007 mosquito season when West Nile virus activity was detected for the first time in two years.

# WNV vector competence for members of the Culex pipiens complex: Cx. p. pipiens, Cx. p. quinquefasciatus, and hybrids

Michael Doyle, mdoyle@cdc.gov, and Harry Savage

Individual adult females reared from field collected egg rafts of *Culex pipiens pipiens*, *Culex p. quinquefasciatus*, and hybrids were infected with WNV by infectious blood meal. The resulting saliva expectorated on day 7-post infection was assayed for quantity of WNV. Preliminary results on variation in WNV vector competence between individuals, maternal families, viral titer at inoculation (i.e., 10^4 or 10^7), and geographic location (i.e., *Culex p. quinquefasciatus* from Louisiana, *Culex p. pipiens* from Chicago, and hybrid specimens from Memphis, Tennessee) will be presented.

# 38 Host choice and WNV infection rates in blood fed mosquitoes, including the *Culex pipiens* complex, from Memphis, Tennessee

Harry M. Savage, hms1@cdc.gov

The source of bloodmeals in 2,082 blood-fed mosquitoes collected from February 2002 through December 2003 in Memphis and surrounding areas of Shelby County, Tennessee were determined. Members of the genus Culex and Anopheles quadrimaculatus predominated in the collections. Members of the Cx. pipiens complex and Cx. restuans were found to feed predominately upon avian hosts, though mammalian hosts made up a substantial proportion of the bloodmeals in these species. No significant difference was seen in the host class of bloodmeals in mosquitoes identified as Cx. pipiens pipiens, Cx. p. auinauefasciatus, or hybrids between these two taxa, Anopheles quadrimaculatus and Cx. erraticus fed primarily upon mammalian hosts. Three avian species (the American robin, the common grackle, and the northern cardinal) made up the majority of avian-derived bloodmeals, with the American robin representing the most frequently fed upon avian host. An analysis of these host-feeding data using a modification of a transmission model for eastern equine encephalitis virus suggested that the American robin and common grackle represented the most important reservoir hosts for West Nile virus. A temporal analysis of the feeding patterns of the dominant Culex species did not support a shift in feeding behavior away from robins to mammals late in the summer. However, a significant degree of temporal variation was noted in the proportion of robin-derived bloodmeals when the data were analyzed by semi-monthly periods throughout the summers of 2002 and 2003. This pattern was consistent with the hypothesis that the mosquitoes were preferentially feeding upon fledglings.

# 39 Highly WNV-endemic sites: The paradox of high infection rates and near-absence of human transmission Jack B. Gingrich, gingrich@udel.edu, Gregory Shriver, and Linda L. O'Connor

We performed a 2-yr longitudinal study of West Nile virus (WNV) infections in wild bird reservoirs, mosquitoes, and sentinel chickens at 6 WNV-endemic sites in DE. *Culex pipiens* was the principal vector at 2 highly endemic sites in 2006 vs. 4 sites in 2007. In 2006, we also found 3 other WNV-positive vector species at 1 site, suggesting increased epidemic potential during 2007. Actual reservoir infection rates were higher during 2007 compared to 2006, as were MFIR's in vector species, even though mosquito-positive numbers declined. Wild birds were early indicators of WNV activity at highly endemic sites in July and early August of both years. During 2006, mosquitoes were positive ~4 wk following evidence of viral circulation in wild birds, appearing concurrently with seroconversion in sentinel chickens. Of birds tested in 2007 (n>8), cardinals had the highest seropositivity rates (53%), followed by house sparrows (29%), Carolina wrens (25%), and American robins (22%). These rates were 2-4 times higher than they were in 2006. However, drought in 2007 probably limited populations of bridge vector species, and therefore limited the disease as an enzootic outbreak. Specific timing of infections in birds, mosquitoes, and sentinels in 2007 is also discussed.

#### 40 Dead birds introduce West Nile virus into residential environments

Carrie Nielsen, Carrie.Nielsen@cdph.ca.gov, and William K. Reisen

A comparative study of West Nile virus (WNV) infection rates in *Culex* mosquitoes collected at 13 sites, 7 reporting WNV-positive dead corvids (case sites) and 6 without reported dead birds (control sites) was conducted in Davis, CA, from 14-21 July at the beginning of the 2006 WNV outbreak. A total of 3,051 *Culex* mosquitoes was collected using gravid traps and CO<sub>2</sub>-baited traps; WNV-infected mosquitoes were only collected with CO<sub>2</sub>-baited traps. WNV-infected *Cx. pipiens* were collected at 1 of the 7 case sites. Six of 7 case sites yielded WNV-infected *Cx. tarsalis*, whereas only 1 of 6 control sites had WNV-infected *Cx. tarsalis*. Overall, the odds of finding WNV-positive mosquitoes were 19.75 times greater at sites reporting a WNV-infected dead corvid than sites without a WNV-infected dead corvid. Maximum likelihood estimates of the overall infection rates at the case sites were 3.48/1,000 for *Cx. tarsalis* and 8.69/1,000 for *Cx. pipiens* compared with 1.02/1,000 in *Cx. tarsalis* collected at the control sites. Results indicate that *Cx. tarsalis* was important in early season enzootic transmission within Davis and that sites reporting WNV-infected dead corvids are areas to focus control and surveillance efforts.

#### 41 West Nile virus activity in CA: An update

Vicki Kramer, vicki.kramer@cdph.ca.gov, and Stan Husted

CA initiated surveillance for West Nile virus (WNV) in 2000 with the first evidence of widespread activity occurring in 2003. In 2004, WNV spread from southern CA to all regions of the state. This presentation will

summarize WNV activity in CA from 2004 through 2007 and discuss the public health impacts of this introduced virus in CA. Steps taken to respond to WNV and reduce the risk of transmission will be summarized.

# 42 Size does matter - effects of size of control area on West Nile human case rates Philip Curry, pcurry@health.gov.sk.ca, Helen Bangura, and Erin Laing

In 2003 and 2007, Saskatchewan and other Great Plains provinces and states experienced severe outbreaks of WNV. In both years, WN virus activity and human disease were more prevalent in rural areas of Saskatchewan, with the southern grassland eco-region having the highest numbers of WN vector mosquitoes, highest amounts of available larval habitat, and warmest temperatures. *Culex tarsalis*, the main vector of the disease in Saskatchewan, is primarily a rural, surface-water, grassland species that migrates into urban areas. Human disease rates and numbers of infected vector mosquitoes were examined in 13 urban communities and in adjacent rural areas within the high WNV risk grassland ecoregion of Saskatchewan. Larger communities that conducted intensive larviciding and source reduction programs within the community and in an extended treatment or buffer area surrounding the community had lower *Cx. tarsalis* numbers in all years. Results were more variable in wet years and were dependent on the amount of habitat available for *Cx. tarsalis*. The size of the community and extended treatment area surrounding the community were related and had an effect on mosquito numbers and disease rates. Human disease rates were lowest in cities with the lowest number of vector mosquitoes and more than four times higher in rural areas.

# 43 Where has all the western equine encephalomyelitis virus gone?

William K. Reisen, arbo123@pacbell.net, Aaron Brault, and Ying Fang

Historically western equine encephalomyelitis virus (WEEV) caused large equine and human epidemics in North America from Canada to Argentina. Despite recent enhanced surveillance for West Nile virus, there have been few reports of equine or human cases and little documented enzootic activity. Recently WEEV has been active in CA, but without human or equine cases. In the current presentation, we compare the host and vector competence of representative WEEV isolates made over the past 60 yrs, using white-crowned sparrows and house sparrows and Culex tarsalis as representative hosts. Results indicate limited variability in virulence among strains in birds and no difference in vector competence. Although genetic changes have been reported, these seem to have limited expression as changes in host competence and cannot explain the absence of equine and horse cases.

# Use of Google Earth for the management of vector-borne diseases in resource-poor environments Darwin Elizondo-Quiroga, darwin\_elizondo@hotmail.com, Saul Lozano-Fuentes, Jose Arturo Farfan-Ale, Maria Alba Loroño-Pino, Julian Garcia-Rejon, Salvador Gomez-Carro, Victor Lira-Zumbardo, Rosario Najera-Vazquez, Ildefonso Fernandez-Salas, Joaquin Calderon-Martinez, Marco Dominguez-Galera, Pedro Mis-Avila, Michael Coleman, Natashia Morris, Chester G. Moore, Barry J. Beaty, and Lars Eisen

The operational value of a decision support system (DSS) for vector-borne disease management is enhanced by a geographic information system (GIS) spatial backbone allowing for visualization of spatiotemporal vector and disease patterns. Resource-poor environments in desperate need of GIS-based solutions for more effective disease management can be faced with the reality that even the most basic GIS data (e.g., administrative boundaries or city structure) are lacking. To address this problem, we have developed methodology for using free mapping tools to generate basic GIS data layers at minimal cost and serve as the spatial underpinning of a DSS for vector-borne disease management. Two cities in Mexico were used to demonstrate that a basic representation of a city useful as the spatial backbone in a dengue DSS can be developed at minimal cost from satellite imagery accessible through a free mapping tool (Google Earth). A stand-alone management tool for extraction of information from a data warehouse and generation of text-format reports and Google Earth-based map outputs also was developed. As a conclusion, the combination of free mapping tools and free or low-cost GIS software has tremendous potential for use in DSS to facilitate control of vector-borne diseases.

### 45 Educating children- a method for controlling Triatomines

José Alejandro Martínez-Ibarra, aibarra@cusur.udg.mx, María Guadalupe Moreno-Ruiz, Héctor Manuel Barajas-Martínez, and Francisco Trujillo-Contreras

In México, control of triatomines mainly relies on the use of insecticides, especially pyrethroids. Besides that method, physical improvement of human dwellings have been carried out in some places of Mexico; however, in some cases, inhabitants of those towns have provided only limited participation, and the populations of bugs has not decreased. Taking in account those considerations, research involving children from 3 to 5 yr old was recently developed in a city in the Jalisco state in western Mexico,. In that place, 2 kindergartens, in 2 similar neighborhoods ("colonias") but far from each other were selected. Entomological indices were recorded before intervention in each study areas. Children in one of the kindergartens were "trained" on Triatominae control by a projection of a serial of images related to triatomines (e.g., their shelters); after that they were trained "in collection of triatomines." Finally, as reinforcement, children were asked to draw those items that were more interesting to them. Meanwhile, the second group was the untreated control. Post-intervention entomological indices were lower in the intervention area than in the control, apparently because the "open mind" of younger children let them receive information more easily.

Ana Soto Vivas, ana.soto@iaesp.edu.ve, Darjaniva Molina de Fernandez, Wilfre Machado, and M. Dora Feliciangeli

An investigation of observational type was designed in 8 villages in Barinas State, Venezuela. The effectiveness of 10% lambdacyhalothrin (WP) 25 mg/m² and fenitrothion 40% (WP), 2 g/m² was measured in function of the mortality of the exposed triatomines using the WHO method. The susceptible "Chagas" strain of *Rhodnius prolixus* was used. Houses were evaluated from the 1<sup>st</sup> day post-spray and then at a monthly frequency. We used the segmentation analysis where the dependent variable was mortality. It showed that the 2 predictors that produced bigger discrimination and that were significant were: Time of action and surfaces. There was 100% mortality for the first 3 days to 30 d post-spray; the surfaces presented mortality of 86.57% at 80 d and the surfaces of wood, cement and painting wattle and daub presented 31.71%. At 90 d the surfaces of wood, cement-lime and wattle and daub-lime presented mortalities between 11.11% and 62.96%. There are no reasons to suppose that the mortality is due to differences in the effectiveness of the insecticides. We demonstrated the interaction between the dependent variable (mortality) and the predictors of time of action and surfaces. There was no significant association in the predictors of temperature and relative humidity with Pearson's correlation (Temperature 0.1580 and Hr -0.0191).

### 47 Community-based entomological surveillance in rural Jalisco, México

Juan I. Arredondo-Jiménez, Ijiarre1@gmail.com, Ezequiel Magallón-Gastélum, Adriana Luna-Zermeño, and Felipe Lozano-Kasten

When asked which household pests were prevalent in the rural municipality of San Martín Hidalgo, Jalisco, México, residents referred to 4 types in the following order: Cockroaches, scorpions, mosquitoes, and kissing bugs. Then, they were asked to collect the bugs by themselves, a task for which they were provided with 4 jars to put each bug type separately. Twenty-two of 23 villages belonging to the municipality, responded and sent samples, all of which were identified to the species level. Out of nearly 1,500 samples analyzed, we found that 90% of all houses had cockroaches (*Periplaneta americana*, *Blatella germanica*), 48% scorpions (mainly *Centruroides noxius*), 83% mosquitoes (*Aedes aegypti*, *Ochlerotatus epactius*, *Culex quinquefasciatus*), and 25% kissing bugs (*Meccus longipennis, Triatoma barberi*). It is worthwhile noting that about 30% of inspected houses were positive for *Ae. aegypti*, indicating that even in this rural area dengue risk is high. It was concluded that community-based entomological surveillance is possible and could help in the monitoring of household bugs, including vectors of disease. This provides the first field example of community based-entomological surveillance in western México.

48 Accurate, rapid and robust sweep-net estimations of total (Diptera: Culicidae) immature numbers Claudia M.E. Romero-Vivas, clromero@uninorte.edu.co, Humberto Llinás, and Andrew K.I. Falconar

The ability of a simple and rapid sweeping method, coupled with calibration 4<sup>th</sup> instar (L3/L4) larvae in large (20 to 6,412 L) water-storage containers at different water levels, in the 19-30° C dengue virus transmission-temperature range, was evaluated. Using this method, 3 calibration factors could accurately and robustly estimate total *Ae. aegypti* pupal numbers in their principal habitat from 14 to 1,630 m above sea-level (19-30° C). New sets of calibration factors were, however, needed to accurately and robustly estimate *Ae. aegypti* L3/L4 larval populations in these water-storage container types at each of 4 study sites at 14, 358, 998 and 1,630 m above sea-level. *Aedes aegypti* L3/L4 larval collections, using this single water surface-layer sweeping method, was less efficient than for pupae despite L3/L4 larvae being present in greater numbers. Because this method was both rapid to perform and did not disturb the sediment layers in these domestic water-storage containers, it was more acceptable by the residents and, therefore, ideally suited for routine surveillance purposes and to assess the efficacy of *Ae. aegypti* control programs in dengue virus-endemic areas throughout the world.

### 49 Altitudinal analysis of dipterans of medical importance in Chipinque Ecological Park

Humberto Quiroz Martínez, hqm\_uanl@yahoo.com, Yuridia Lisseth González López, Adriana Rodríguez Castro, Eric Iván Meléndez López, and Eduardo Alanis Rodríguez

A study of Diptera diversity at different elevations was conducted in the Chipinque Ecological Park. Ovitraps were used to collect mosquitoes at 6 sampling sites from 900 to 1,280 m above sea level. Samples were taken on a weekly and later identified in the laboratory using keys by Merrit and Cummins (1996) and Darsie and Ward (1981). Diptera collected were Culicidae and Ephydridae with *Culex tarsalis*, *Cx. restuans*, *Aedes abserratus* and *Toxorhynchites* spp.

# Multipurpose indoor residual spray of lambdacyhalothrin (10WP) to control household pests in rural Jalisco,

Juan I. Arredondo-Jiménez, jiarrel@gmail.com, Adriana Luna-Zermeño, Manuel Raygoza, and Felipe Lozano-Kasten

We investigated the effect of indoor residual spray of lambdacyhalothrin against household pests (cockroaches, scorpions, mosquitoes and kissing bugs) in rural Jalisco, México. Following the community practice approach, we trained and supervised local spraymen according to standard protocols. The first time in Mexico, funds for the insecticide and its application were covered by the municipal authority and residents. Insecticide was applied at 20 mg ai/m² considering that over 90% of wall surfaces were very porous, unplastered bricks. Residual life of the insecticide was assessed with monthly wall bioassays using *Aedes aegypti*. Household interviews and house inspections were carried out to assess insecticide impact on cockroaches, scorpions and kissing bugs. Because we

undertook all measures to reduce acute exposure to insecticide, we heard no complaints after its application by residents or spraymen. It was found that the insecticide remained effective for up to 12 wk according to the mosquito wall bioassays. It was also found that infestations by other bugs remained very low over the same period of time. We concluded that insecticide treatments can be conducted by properly supervised local spraymen, that lambdacyhalothrin was effective against all intended targets and that insecticide re-applications were welcomed by residents even when they had to be paid for.

### 51 Efficient protein extraction in *Aedes aegypti* for proteome analysis

Flor Herrera, flormhq@gmail.com, Jose Rivero, Susana Zahalan, Nardy Diez, and Yasmin Rubio-Palis

Insecticide resistance has a negative impact on the control of mosquito-borne diseases like dengue. Application of proteomics and protein identification methods together with the completion of genome sequences for insects give us the tools for examining insect resistance at a molecular level. We are interested in studying the proteins of *Aedes aegypti* from different regions of Venezuela with variable degrees of insecticide resistance. For this, we extracted proteins from *Ae. aegypti* using Triton X-114 (Bordier C., JBC 1980, 256: 1604-1607) to solubilize membranes and whole cells and to separate integral membrane proteins from hydrophilic proteins. Results showed that the quantity of proteins obtained was 8-10% of the total mosquito weight. The distribution of the proteins was 90% hydrophilic and 10% hydrophobic. The proteins were then separated by bidimensional electrophoresis revealing the presence of approximately 60 and 30 spots for the hydrophilic and hydrophobic proteins, respectively. Financial support was provided by CDCH-UC and by the FONACIT, Venezuela.

### 52 Dry season investigation of Aedes aegypti in a dengue endemic area

Roberto Barrera, rbarrera@cdc.gov, Mary Hayden, Manuel Amador, Annette Diaz, and Joshua Smith

We investigated *Aedes aegypti* (pupal survey, adults) in 2 neighborhoods with high (H) and low (L) dengue endemicity during the dry season of 2007 in San Juan, Puerto Rico, (March – April) to determine if dengue endemicity was associated with the permanency of aquatic habitats and mosquito productivity. Our hypothesis was that dengue endemicity is due to the presence of containers with water during the low rainfall season that are generated by humans (water storage vessels), climate (rainfall uniformly distributed in time), or both. We sampled mosquito pupae in 300 houses in each neighborhood. Adult mosquitoes were captured using 20 lured BG-sentinel traps for 2 wk (160 trap-days). We sequentially sampled the H neighborhood for 2 weeks 1<sup>st</sup>, then the L neighborhood for another 2 wk. Heavy rainfall fell for 1 d during the 2<sup>nd</sup> wk of the study, possibly affecting our measures of mosquito abundance in L. More pupae and pupae per container of *Ae. aegypti* were found in H (1,384;  $2.2 \pm 0.5$ ) than in L (909;  $1.1 \pm 0.2$ ). The mean number of *Ae. aegypti* pupae in householder water-filled containers in H ( $2.9 \pm 0.7$ ) was larger than in L ( $0.8 \pm 0.2$ ), but the reverse was observed in rain water-filled containers in H ( $0.7 \pm 0.3$ ) and L ( $0.7 \pm 0.7$ ). Significanly more adult *Ae. aegypti* were captured in L (0.633 females, 0.745 males) than in H (0.755 females, 0.755 fe

### 53 The use of ovitraps index to monitor Aedes aegypti in Belo Horizonte, Brasil

Adelaide Maria Sales Bessa; Simone Costa Araújo; Jose Carlos Nascimento; Fernanda Carvalho De Menezes; José Eduardo Pessanha; Ione Oliveira Costa, entolzoon@pbh.gov.br

Since 2001, the Zoonosis Control Department (ZCD) in Belo Horizonte (BH) has subsidized vector control activities by providing ovitraps. The ZCD verified that this practice is very useful in the detection of new infestations and in monitoring *Aedes aegypti* adult populations. We analyzed data from January 2003 to December 2006. BH possesses 1,686 points where ovitraps are distributed in the nine regions of the municipality. These traps are operated biweekly, maintaining rays of 200m. The 144,471 traps were operated and 47,721 presented eggs. The index of positivity of ovitraps (IPO) and the index of density of eggs (IDO) were 33.03 and 58.68, respectively. The IDO varied by yr from 55.42 to 69.11 and by week from 82.69 to 24.34, indicating great variation during the year. We observed greater predominance of *Ae. aegypti* than *Ae. albopictus* during the study. This method proved to be sensitive and economical in the detection of *Ae. aegypti*. In periods of low infestation where larvae surveillance is less effective, the traps aided in a decisive way in the early discovery of new foci. The data generated support, in a routine way, the vector control and reinforce the efficiency of the method.

# 54 Laboratory evaluation of spatial repellency and irritability of permethrin for *Aedes aegypti* (L.) Rocio Ramirez Jimenez, rociormz14@hotmail.com, Ewry Zarate-Nahon, A. E. Flores, and Idelfonso Fernandez Salas

Dengue, a major public health problem throughout subtropical and tropical regions, is an acute infectious disease characterized by biphasic fever, headache, pain in various parts of the body, rash, lymphadenopathy, and leukopenia. *Aedes aegypti* (L.), the primary vector of dengue and urban yellow fever, exhibits resistance to several insecticides, a situation that creates many problems for vector control programs in several countries. In the present research, the HITSS system was used; this system has a modular design that examines several aspects (contact irritation, spatial repellency and toxicity). Currently, there are few tests for evaluating contact irritation and spatial repellency that are produced by adulticides on mosquito behavior with the exception of the test for penetrating response. Still, there is no standard system for assessing new chemical agents that affect adult mosquito behavior. The behavioral response of *Ae. aegypti* to several doses (0.025nmol/cm², 0.25 nmol/cm², 2.5 nmol/cm² and 25 nmol/cm²) of the topical insecticide permethrin was evaluated. At a concentration of 25 nmol/cm², 51.6% contact

irritation was obtained; at 25 nmol/cm<sup>2</sup>, 56.66% toxicity was observed; and for spatial repellency, the response was similar at all doses.

# 55 Synergistic effect of s,s,s,-tributylphosphorotrithioate (DEF), piperonyl butoxide (PB) and permethrin in two subpopulations of *Aedes aegypti* from northeastern Mexico

A. E. Flores, adrflores@gmail.com, Carlos H. Marin Hernandez, Juan F. Martínez Perales, Beatriz Lopez Monroy, Antonio Cortes Guzman, Gustavo Ponce Garcia, and Ildefonso Fernandez Salas

The bottle bioassay was utilized to determine the modulating effect of S,S,S,-tributylphosphorotrithioate (DEF) or piperonyl butoxide (PB) in combination with the pyrethroid insecticide permethrin on F1 adult females of 2 subpopulations of *Aedes aegypti* collected in the metropolitan area of Monterrey, Nuevo Leon, Mexico. The optimal dose of each synergist was determined as the maximal dose that did not cause mortality. The results obtained from the bioassays were analyzed to determine in each population the  $LC_{50}$  of the insecticide alone and in combination with each of the synergists, as well as the toxicity ratio (TRx). The  $LC_{50}$  of permethrin of subpopulation 1 was 0.45 ppm (0.30-0.65 ppm); while for the combination with DEF, it was 0.032 ppm (0.014-0.060 ppm) with a TR of 14.06X. For subpopulation 2, the  $LC_{50}$  of permethrin was 1.92 ppm (1.57-2.31 ppm), and for the combination with PB 0.31 ppm (0.21-0.48 ppm) with a TR of 6.19X. The magnitude of the TR indicates that esterases are important in the detoxification of permethrin and that the synergist DEF could be a good choice for formulating with permethrin.

### 56 Is it globalization or climate change?

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The escalating mobility of exotic vectors and pathogens is a major cause for concern. A classic example is the spectacular rate with which *Aedes albopictus* has colonized the Americas, Europe and Africa, and its recent role as the epidemic vector of chikungunya virus in Italy. It is clearly misleading to state that these events are the result of climate change, yet the public is repeatedly bombarded with such claims from "experts", including some from the world's most prestigious institutions. It is our duty as scientists to defend our science by correcting such misinformation.

### 57 Anatomy of an epidemic: Movement of chikungunya virus

Ann Powers, APowers@cdc.gov

One alphavirus, chikungunya virus (CHIKV), has been reported to be the cause of recent explosive outbreaks that have been associated with severe morbidity. Traditionally, CHIKV causes a febrile illness where the characteristic feature is a debilitating and prolonged arthralgic syndrome that primarily affects the peripheral small joints. The pain associated with CHIKV infection of the joints typically persists for weeks or months. During the 2004-2007 ongoing explosive epidemics on the Indian Ocean islands and India, suggested changes in transmission patterns, distribution, and epidemiology have been recorded. A discussion of clinical presentation, virus-vector associations, and genetics of the virus over time to assess the possibility of further expansion and re-emergence of CHIKV illness will be presented.

#### 58 Potential for dengue transmission in the continental US

Gary Clark, gary.clark@ars.usda.gov

This presentation describes the transmission cycle, epidemiology and recent incidence of dengue in the Americas. Critical factors, including the characteristics of competent mosquito vectors, frequent introductions of dengue viruses into the US, and a highly susceptible human population that are needed for indigenous transmission, will be reviewed. Factors mitigating possible transmission and a best case/worst case scenario for indigenous transmission to occur will also be presented. Recommendations for detecting and responding to indigenous dengue transmission will be made. Data on the occurrence of dengue cases detected in the US since 1960 will be presented along with data from Texas from 1945 to date. The presentation will conclude with information about two recent transmission episodes along the Texas-Mexico border and their implications.

### 59 Potential for Rift Valley to be introduced into North America

Kenneth Linthicum, Kenneth.Linthicum@ars.usda.gov, Assaf Anyamba, Jennifer Small, Compton Tucker, Jean-Paul Chretien, and Seth Britch

Rift Valley fever (RVF) is a mosquito-borne zoonotic disease of domestic ruminants in Africa. The disease is most severe in cattle, sheep, and goats, causing mortality in young animals and abortion in adults. Human infection causes significant morbidity and mortality. RVF occurs in sub-Saharan Africa. The disease first spread outside sub-Saharan Africa into Egypt in 1997 and resulted in large losses among the domestic animal populations and caused significant human disease. Subsequently, in 1987, a large outbreak in animals and people occurred in Sahel region of Senegal and Mauritania, and then in September 2000, a RVF outbreak occurred in Saudi Arabia and Yemen along the Red Sea Coast, representing the first RVF cases identified outside Africa. After the virus is introduced into domestic animals, a wide variety of mosquito species may serve as a vector. There are no licensed animal or human vaccines available for use in the United States, and there is minimal surveillance for the disease in North America. Here we discuss the potential for the disease to be introduced into North America, and strategies to (1) prevent its introduction, (2) detect its introduction, and (3) eradicate it should it be introduced.

# 60 Culicoides-borne emerging pathogens: The reemergence of bluetongue virus Walter Tabachnick, wjt@ufl.edu

Different species in the genus *Culicoides* transmit animal pathogens such as bluetongue, epizootic hemorrhagic disease, and African horse sickness viruses. The distribution of these viruses is limited to geographic areas containing competent vector species. Bluetongue virus illustrates the potential for the rapid re-emergence of *Culicoides*-borne animal pathogens, which could have devastating consequences on the livestock economies of many countries including the US. There are several different epidemiologic systems in the world with different species of *Culicoides* responsible for transmitting specific serotypes of bluetongue virus. The recent spread of bluetongue virus to northern Europe and the United Kingdom demonstrates the danger of the introduction of new serotypes into new epidemiological systems. Our ability to predict and/or mitigate bluetongue in different regions will require a substantial investment in new research paradigms that investigate details of underlying controlling mechanisms in several species of *Culicoides*. The epidemiology of bluetongue is well characterized in the US, compared to other regions of the world. Despite this, the introduction of exotic bluetongue viruses into the US could change the current situation and poses a real risk to the health of US livestock.

# 61 Potential for transmission of Venezuelan equine encephalomyelitis or Japanese encephalitis viruses in the continental United States

Michael Turell, Michael.turell@amedd.army.mil

The introduction and spread of West Nile virus in the Americas illustrates the potential for an exotic arthropod-borne pathogen to become established in North America. Other viruses such as Venezuelan equine encephalomyelitis virus (VEEV) and Japanese encephalitis virus (JEV) have the potential to become established in North America and cause significant disease in both humans and domestic animals. Infection with VEEV, an alphavirus in the family Togaviridae, in humans usually results in febrile illness and occasionally encephalitis and death, while infections in equines are often fatal. JEV, a flavivirus in the family Flaviviridae, is responsible for thousands of encephalitis deaths in Asia and India each year. While Ochlerotatus taeniorhynchus has been incriminated as the principal epizootic vector of VEEV, various members of the subgenus Culex (Melanoconion) may be involved in the enzootic maintenance of these viruses. In contrast, Cx. (Culex) tritaeniorhynchus is considered the principal vector of JEV. Because mosquito control methods are often species specific, it is important to determine which mosquito species are capable of transmitting these viruses so that appropriate control measures can be instituted rapidly should either of these viruses be reintroduced into the United States.

#### 62 Introduction

Roger Nasci, RNasci@cdc.gov

Hurricanes Katrina and Rita had impacts on both mosquitoes and mosquito control programs in AL, MS, LA, and TX. The US Congress recognized the need to address mosquito control in these areas and provided funds through a supplemental appropriations act in 2006 to support and enhance mosquito control programs in communities affected by the storms. The CDC was charged with developing a plan to distribute the funds, and with evaluating the effectiveness of the program. In this symposium, each of the states receiving funds will report the effect of the storms on mosquitoes and mosquito control, how each state assessed needs and distributed support, and how the program influenced the expansion and enhancement of mosquito control.

#### 63 Supplemental mosquito control program in AL

Julius Grady, fredgrady@adph.state.al.us, C. F. Erdman, Zen Yue, Jackie Holliday, and Ruth Underwood

After the notice of award for Mosquito Abatement and Control in the Katrina affected areas of AL, staff from the named counties, area health offices, and the State Department of Public Health Central Office began evaluating the communities' risks and needs. Local public health staff (environmentalist and health administrators) facilitated meetings and helped with the drafting of the local submissions. Proposals were received from towns, cities, and counties. These proposals were reviewed and the awards were announced. Counties then agreed upon contracts to accomplish their goals and objectives specified in their proposed mosquito abatement plan. Each project was required first, to begin with a pre-intervention report to include their existing problems and proposed activities; second, to document the progress of their projects; and finally, to submit a post-project report including the objectives met and unmet. AL was late receiving their funds and most of the activity occurred in 2007.

### 64 Supplemental mosquito control program in MS

Kristine Edwards, msmosquitoes@bellsouth.net, Jerome Goddard, and Wendy Varnado

In 2006, MS received approximately \$2.8 million through a supplemental appropriation by the US Congress in order to support and enhance mosquito control in communities affected by Hurricane Katrina. The MS Department of Health (MDH) distributed the funds by issuing a request for proposals to all counties and municipalities in the 49 counties declared disaster areas. Proposals were scored on a scale by a panel of experts at the MDH; preference was given to proposals which included integrated mosquito control methods. Qualifying communities were then issued contracts for disbursement of the funds. Only mosquito control equipment, pesticides, and fuel/labor (for mosquito control) were reimbursed under terms of the contract. Eighty-seven contracts were issued. A survey of mosquito control personnel was conducted to evaluate effectiveness of mosquito control programs at these 87 sites. The survey (a questionnaire) explored the status of each mosquito control program before awarding of funds, and then

another questionnaire after funding was designed to assess changes in procedures, as well as changes in attitudes regarding mosquito control. Results of these questionnaires will be presented and implications discussed.

# 65 Supplemental mosquito control program in LA

Kyle Moppert, KMoppert@dhh.la.gov

In 2005, both Hurricane Katrina and Rita made landfall in LA. Their related storm surges and levee failures flooded historically large urban and rural regions with significant impacts on both mosquitoes and mosquito control programs across south LA. Impacts to mosquito control programs ranged from total destruction to increased cost of providing protection to large populations living without power and subject to extremely high populations of storm related mosquitoes. The \$3.2 million dollars received from the supplemental appropriations act in 2006 enabled the LA Department of Health and Hospitals - Office of Public Health – to assess the needs of and distribute support to 26 mosquito control programs across the state. A weighted system of distribution allowed those programs, which were impacted most, to receive higher funding, while still assisting all qualifying surveillance based mosquito control programs. Funding ranged from \$80,057 to \$320,228. In addition, these funds enabled the expansion and enhancement of several previously non-qualifying mosquito control programs to become surveillance-based programs.

### 66 Supplemental mosquito control program in TX

Roy Burton, Roy.Burton@dshs.state.tx.us, and Jeff Taylor

TX Department of State Health Services (DSHS) received \$1,108,618 to assist 22 counties in southeast TX adversely affected by Hurricane Rita. A work group was created to provide guidance to DSHS in allocation and use of the funds. Allocation of funds was based on (1) existence of an established mosquito control program; (2) Hurricane Rita impact level; (3) documented mosquito landing rate counts, and (4) occurrence of West Nile virus. DSHS mailed letters to the county judges in these 22 counties offering funds for mosquito control activities. The letter also provided a checklist of authorized purchases and a checklist of integrated mosquito management activities. DSHS subsequently developed contracts with 10 counties.

#### 67 Welcome to the Trustee/Commissioner Session

Mike Morris, Northern Salinas Valley MAD, Salinas, CA

Using a variety of speakers with local, state, and federal expertise, this Trustee Session will provide an overview of legal, financial, and lobbying issues with mosquito control operations should consider. This session will also include several papers providing perspective on the efficacy of larviciding vs. adulticiding and considerations concerning ground vs. aerial adulticiding.

# 68 Building relationships with your local elected officials and agency personnel Ralph A. Heim, (916) 441-0702 email?

It is critical to build relationships with your local elected officials and state agency personnel. These relationships and effective communication are among the key components for successful lobbying of these decision makers.

# 69 The Washington scene: The AMCA's lobbying efforts in Washington

Ed Ruckert, eruckert@mwe.com

The AMCA's lobbyist in Washington will provide an overview of the political situation in Washington as it relates to mosquito control interests and include an update on legislative and regulatory issues of importance to our industry.

### 70 Impacts of environmental regulation and issues on mosquito and vector control

Richard Shanahan, rps@bkslawfirm.com

This presentation will focus on significant nationwide issues involving environmental law and regulation that affect mosquito and vector control and current litigation of national interest.

# 71 Gaining financial independence for your district: Funding mosquito control services

Brian Jewett, brianj@muni.com

State and county funding for mosquito abatement districts can vary from year to year. This presentation will examine additional funding sources for districts to help stabilize their revenue such as benefit assessment districts, parcel taxes, user fees, and other revenue sources.

# 72 Understanding stormwater BMP and their unintended impact on mosquito control agencies Marco Metzger, Marco.Metzger@cdph.ca.gov

Federal, state, and local clean water regulations are forcing tremendous changes in the treatment and management of urban and stormwater runoff. Implementation of structural treatment devices known as best management practices (BMP) is required under existing laws in an effort to improve the quality of water runoff before it enters receiving waters. An unintended consequence of BMP implementation is the creation of mosquito habitats when not properly designed and/or maintained. Since 1999, the CA Department of Public Health, Vector-Borne Disease

Section has led a series of collaborative studies to identify conditions conducive to mosquito production in BMP and develop long-term mosquito management solutions for these structures. The results of these efforts have contributed greatly to how subsequent BMP have been designed, implemented, and maintained; however, many critical problems remain unresolved. This presentation will provide an introduction and overview of stormwater runoff management, the associated vector issues, measures for successfully managing mosquitoes, and existing and future challenges.

73 Evaluation of copepods for the control of *Aedes aegypti* (Diptera: Culicidae) in northern Colombia Claudia M. E. Romero-Vivas, clromero@uninorte.edu.co, Luis Eduardo Castro-Cera, and Andrew K.I. Falconar

Different species of copepods were collected from various types of natural fresh water bodies in Departamento del Atlántico (Colombia) to identify those with the highest predatory capacity for *Aedes aegypti* larvae in the laboratory and under semi-field conditions. Although we found 3 species already known for their capacity of controlling mosquito vectors: *Mesocyclops brasilianus*, *M. aspericornis* and *M. meridianus*, we also found 6 other species within this order about which little is known concerning their ecology and geographic distribution. The predatory capacities of these species were determined in the laboratory using Petri dishes of 55 mm in diameter and 14 mm high, with 20 ml of dechlorinated water. Ten replicates of different numbers of L-1 larvae per copepod were used: 5, 10, 20, 30, 40 and 50. Before each test, the copepods were left without food for 72 h. There were significant differences in the average number of larvae attacked (df1= 2, df2= 172, F=68,4 y p <0,05) with *M. meridianus* as the species with the highest capacity (79%) followed by *M. aspericornis* (68%) and *M. brasilianus* (37%). *Mesocyclops aspericornis* were applied in the field and currently are being evaluated. Results of the field studies will be discussed.

74 Evaluation of larvicidal activity of Eupatorium microphyllum extract against Aedes aegypti Alvaro Rozo, arozo@lasalle.edu.co, Cristina Zapata, and Felio Bello

The objective of this study was to evaluate the larvicidal activity of crude acetone and watery extracts of the plant *Eupatorium microphyllum* against *Aedes aegypti* 4<sup>th</sup>-instar larvae. Crude watery extracts were used in concentrations of 5, 15, and 25% and crude acetone extracts were used in concentrations 0.1, 0.2, 0.3, 0.4, and 0.5%. In each bioassay, we placed 20 larvae in plastic cups with 150 ml of watery and acetone extracts. Every bioassay and its corresponding control were carried out in triplicate. Fifty mg/l of *E. microphyllum* acetone extract showed the highest toxic activity on larvae in 24 h under laboratory conditions (95.4% mortality), whereas with watery extract the larval mortality was less than 20%.

75 Improving specifications and evaluation of space spraying equipment for *Aedes aegypti* control in Brasil Paulo César da Silva, paulo.cesar@saude.gov.br, and João Pimenta

Part of the responsibility of the Ministry of Health (MOH) of Brasil is to strengthen the operational capacity of the states and municipalities in Brasil to control *Aedes aegypti* and prevent dengue outbreaks. Because of the demand for space spraying equipment and the difficulties encountered in their selection and purchase, the MOH has initiated a technical program to prepare specifications and evaluate prospective equipment with the objective of improving the quality and durability of the truck-mounted and portable ULV equipment. The MOH has entered into a technical agreement with the engineering department of the University of Brasilia to develop the specifications and conduct the evaluation based a 20-yr performance history of equipment utilized by the national dengue control program. During the initial 18-mo period, special equipment was developed to measure and monitor performance specifications so that detailed specifications could be developed to guide and assist in the evaluation of equipment. These were announced in the official daily government publication and published on the MOH webpage. Upon completion of this initial phase, the MOH is open to discussions and suggestions with industry to improve the guidelines and specifications.

76 Permethrin and temephos resistance in five populations of *Aedes aegypti* (L.) in southern Mexico Brenda G. Silva Salinas, silvaqbp@gmail.com, A. E. Flores, Dulce L. Tovar Martinez, Addie Z. Gutierrez Garcia, Gustavo Ponce Garcia, Ildefonso Fernandez Salas, and William C. Black IV

Five field populations of *Aedes aegypti* were collected in Quintana Roo, México. Ovitraps were set in 3 different places (Lagunitas, Antorchista and Calderitas) in Chetumal and in 2 other municipalities (Solidaridad and Lazaro Cardenas) in Quintana Roo. F1 larvae and adult mosquitoes were tested for susceptibility based on LC-50 and LC-95 against temephos (98% CHEM Service, West Chester, PA) and permethrin (98% CHEM Service, West Chester, PA), the most common insecticides used for dengue control programs in Mexico. Resistance ratio (RR) values for temephos in *Ae. aegypti* populations indicated that populations of Calderitas (RR= 82X) and Lagunitas (59X) are highly resistant to this insecticide compared with the susceptible New Orleans strain. Other populations studied did not show resistance to temephos. Analyzing resistance enzymes, we found iAChE (insensitive acetylcholinesterase) as the main mechanism of temephos resistance. In contrast to temephos results, all populations studied showed low RR values for permethrin, indicating that we do not yet have resistance to this insecticide in these populations.

# 77 Insecticide resistance mechanisms in Aedes aegypti (L.) from Mexico 2002-2007

A. E. Flores, adrflores@gmail.com, Gustavo Ponce Garcia, and Ildefonso Fernandez Salas

We have used existing bioassays and biochemical assays to identify and document mechanisms of resistance of *Aedes aegypti* populations in Mexico. This was done to provide baseline data for program planning and pesticide selection for Mexico. Baseline information was collected on the susceptibility of larval and adult populations of *Ae. aegypti* to the most commonly used pesticides: temephos and permethrin. The results indicate that esterases account for the main resistance mechanisms in permethrin-selected populations, and less frequently involved are glutathione-S-transferase (GST) and oxidases (MFO). In larval populations, mechanisms related to GST and altered acetylcholinesterase (iAChE) appeared to be the principal ones associated with survival in bioassays.

### 78 Resistance mechanisms to temephos in a population of Aedes aegypti from Peru

O. Sarai Sanchez-Rodriguez, saro\_84@hotmail.com, A. E. Flores, Nydia A. Rodriguez-Neavez, Gustavo Ponce-García, Ildefonso Fernandez-Salas, and William C. Black IV

A population of *Aedes aegypti* from Peru was studied for its susceptibility to the larvicide temephos. The  $LC_{50}$  was calculated based on a 24-h exposure of late  $3^{rd}$  and early  $4^{th}$  instar larvae. The dead and living individuals were separated after exposure to the  $LC_{50}$  and microassays were used to determine the activities of detoxifying enzymes including: a esterases,  $\beta$  esterases, oxidases (MFO), glutathione-S-transferase (GST), acetylcholinesterase (AChE) and insensitive acetylcholinesterase (iAChE). The results were compared with those obtained for the susceptible New Orleans (NO) strain which was studied in the same manner as the Peruvian population. The  $LC_{50}$  of temephos for the population from Peru was  $0.034~\mu$ g/mL and the resistance ratio based on the  $LC_{50}$  of the susceptible NO strain was 13.76X. The enzymes that surpassed the tolerance threshold established by NO strain were a esterases with 21.6% and GST with 96.67% (p<0.05).

79 Use of simulated field studies to characterize the development of resistance to larvicides in *Aedes aegypti* Ima Aparecida Braga, ima.braga@saude.gov.br, Denise Valle, and José Bento Pereira Lima

The Brasilian network to monitor *Aedes aegypti* resistance to insecticides (MoReNAa) is conducting studies to understand the dynamics and effects of field conditions on residual larvacides. In Brasil, several types of containers are used domestically to store water and have been shown to be important sources for *Ae, aegypti*. Because of constant use, water levels and the amount of insecticide change frequently in these containers. To determine the persistence of temephos and other larvicides on resistant and susceptible (non-resistant) populations, *Ae. aegypti* were exposed to field-simulated bioassays. These bioassays were conducted both indoors and outdoors. In each milieu, 3 randomly dispersed 50-L containers filled with tap water were used for each population under test and for the Rockefeller strain. On the 1<sup>st</sup> day, each container received the amount of insecticide recommended for field use by the National Dengue Program. Three additional containers remained without temephos treatment. Three times a wk, 10 L of water from each container was replaced with fresh water. Weekly, 50 L3 larvae of the corresponding populations were added to each container, and mortality was recorded 24 and 48 h later, when remaining larvae were discarded. The results of these studies with larvicides in Brasil will be presented.

# 80 Entomological and social evaluation of dengue transmission in areas of high and low disease incidence in Medellín and Bello, Colombia

Liliana P. Elorza-Vélez, lilielorza@yahoo.es, José Azoh, Mario Lamfri, and Guillermo L. Rúa-Uribe (presenter), Grupo de Entomologia Medica (GEM), Universidad de Antioquia, Medellin, Colombia

Dengue is the main vector-borne viral disease in Colombia. Its transmission is determined mainly by the vector mosquito population and by dynamic, socioeconomic and climatic conditions. The interaction of these factors leads to differences in disease incidence among localities. In this study, *Aedes* infestation indices were compared among localities of high and low incidence in Medellín and Bello. Productivity by container type was estimated and a knowledge, attitudes, practices and beliefs (KAPB) survey about dengue prevention was conducted; 597 houses were visited and 5,557 containers were inspected. Entomological transmission risk was observed in high incidence localities as well as in low incidence ones. When areas of high incidence were compared, Bello displayed higher values in entomological indices in contrast to what was observed in Medellín. The discarded tires proved to be the most productive habitats. The social analysis performed found that 80% of study participants know that dengue was transmitted by *Ae. aegypti*, however, only 65% could identify containers as larval habitats. Research findings demonstrated that a close relationship between entomological indices and disease incidence is not occurring. This confirms that more accurate entomological indicators for dengue are needed. This study was supported by IAI (TISG.J2), COLCIENCIAS and Universidad de Antioquia.

81 Evidence of vertical transmission of dengue virus in Aedes albopictus and Ae. aegypti in southeastern Mexico Rogelio Danis Lozano, rdanis@insp.mx, Iliana Rosalía Malo, Mario Henry Rodríguez López, Armando Ulloa García, Celso Ramos García, Lilia Juarez Palma, Ramón Solís, Rosa Maza, Ricardo Nagaya Escobar, Rafael Vazquez Sánchez, and Luis Manuel Salgado Corsantes

The potential of vertical transmission of dengue virus in *Aedes albopictus* and *Ae. aegypti* was explored in 3 endemic neighborhoods in Tapachula municipality in southeastern Chiapas state during a dengue outbreak in 2006. In this outbreak, there were 85 reported cases, of which we studied 48 cases of dengue classic and 6 cases of dengue hemorrhagic fever which were detected by the Ministry of Health's epidemiological surveillance system. In

each neighborhood, a random buffer of 200 m was randomly selected between 30 and 24 houses. Two plastic cans were placed in each house which were changed every 5 d; positive cans were recorded and transferred to the insectary for emergence. The adult mosquitoes which emerged were groups of pools with 1 to 25 females and analyzed by reverse transcription-polymerase chain reaction (RT-PCR) technique to detect the dengue viral genome. The average number of eggs recorded inside and outside houses was 9.13 and 5.30, respectively; 5/550 pools were positive for dengue viruses. We found vertical transmission in *Ae. albopictus* in 1 neighborhood from Tapachula with serotypes DEN-1 and DEN-2 and in another neighborhood we found serotype DEN-3. *Aedes aegypti* was found in 10 neighborhoods of the study area mainly in the center of the community. Vertical transmission was found in *Ae. albopictus* and *Ae. aegypti* in Chiapas.

# 82 FourStar<sup>TM</sup> microbial briquet efficacy in highly organic catch basin microcosms Bob Sjogren, bsjogren@gmail.com, and David Sjogren

FourStar<sup>TM</sup> microbial (*Bacillus sphaericus & Bacillus thuringiensis israelensis*) sustained release briquets provided long lasting mosquito larval control in replicated, highly organic simulated street catch basin microcosms. Fifteen and 30-g briquet trials provided >98% control within 7 d, and for durations of >40 and >120 d, respectively. Mosquito larvae developed up to 3<sup>rd</sup> instar, at which time most died. A few survived to die in 4<sup>th</sup> instar. Almost none developed to pupae.

# 83 FourStar<sup>TM</sup> microbial briquet efficacy in abandoned swimming pools

Bob Sjogren, bsjogren@gmail.com, Ron Berg, Rick Albrecht, Stacy Toynbee, and Debbie Mundeen

FourStar<sup>TM</sup> microbial (*Bacillus sphaericus & Bacillus thuringiensis israelensis*) sustained release briquets were evaluated in abandoned swimming pools. Application rates of 1/100 ft<sup>2</sup> of pool surface controlled larvae within 7 d and prevented pupal development by >98% for >100 d. Mosquito larvae developed up to 3<sup>rd</sup> instar at which time most larvae died. A few survived to die in 4<sup>th</sup> instar stage, almost none developed to pupae.

# 84 Impact of heat during handling and storage on the activities of mosquito larvicides

T. Steven Su, tsu@wvmvcd.org, and Min-Lee Cheng

Larviciding is one of the most cost-effective means for mosquito control. The biorational larvicides, such as *Bti*, *B. sphaericus*, and IGR-based larvicides are the most widely used mosquito larvicides today because their specificity and effectiveness on target organisms and compatibility with the environment. By using standard laboratory bioassay techniques, we have examined the stability of larvicidal activities as a function of temperature of several formulations of *Bti*, *B. sphaericus*, and IGR larvicides. Based on the assay results, we are making recommendations for proper inventory control, handling, and storage of larvicides to minimize loss of larvicidal effectiveness.

# 85 Environmental concentrations of encapsulated methoprene (Altosid-SR) in salt marshes: Measurements and implications

David J. Tonjes, david.tonjes@stonybrook.edu, and Bruce Brownawell

In conjunction with a project to revise and optimize a mosquito control program for Suffolk County (Long Island), NY, operational applications of methoprene (delivered by helicopter as Altosid-SR) were monitored in mosquito control ditches in 2 salt marshes. Some of the aqueous samples were analyzed using high performance liquid chromatography coupled to time-of-flight mass spectroscopy (LC-TOF-MS) by Stony Brook, and others were analyzed by the US Geological Survey using gas chromatography quadruple mass spectrometry (GC-MS). Sediment samples were analyzed by Stony Brook using GC-MS. The data show that methoprene concentrations, while initially exceeding 1 part per billion (ppb) immediately after applications, quickly decrease to part-per-trillion levels, which are sometimes measurable a week or more after the application. Despite weekly reapplications, sediment concentrations do not show any increases in the pesticide load, and did not exceed 100 ppb. The data suggest non-target impacts from methoprene are unlikely to occur to water column aquatic organisms, but also suggest the time-release formulations of Altosid may not maintain concentrations sufficient to kill mosquitoes except immediately after applications.

# 86 Evaluation of Altosid® XR-G sand for expansion of control of *Coquillettidia perturbans* mosquitoes in MN Mark E. Smith, mmcd\_mes@mmcd.org, and Stephen Manweiler

The Metropolitan Mosquito Control District evaluated Altosid<sup>®</sup> XR-G sand to control *Coquillettidia perturbans* mosquitoes in cattail habitat. This evaluation compared Altosid<sup>®</sup> XR-G to Altosid<sup>®</sup> pellet applications and untreated controls. Other aspects reviewed were application issues, swath coverage, and prioritization of helicopter resources. Preliminary results suggest that adding Altosid<sup>®</sup> XR-G will enable us to expand our *Cq. perturbans* control program.

# 87 Evaluation of acoustic larvicide in wastewater treatment plants

Herbert Nyberg, info@larvasonic.com

Acoustic larvicide is the process of killing mosquito larvae in water by introducing acoustic energy at the resonant frequency of the air in their abdomen. New Mountain Innovations conducted surveys and tests in a wastewater treatment plant. Acoustic larvicide equipment was permanently installed in one area of the plant to determine efficacy. The equipment demonstrated 100% control in the area in which it was installed. Larval surveys

determined specific migration patterns within the plant. These patterns can be used to economically install future equipment.

# 88 AGNIQUE® MMF G: Physical properties, active release, and bioefficacy

Dean A. Oester, dean.oester@cognis.com, and Alefesh Hailu

Monomolecular surface films in a solid dosage form have until recently not been commercially available. The technical challenge of loading a sufficiently high percentage of a liquid substance into a solid-dose form that can be applied at typical granule application rates for mosquito control has now been accomplished. AGNIQUE® MMF has been incorporated into a solid form at a concentration of 32%. The physical properties of the granule and the release of the active ingredient from the solid matrix can be adjusted with modifications in the formulation. When monomolecular film formation is followed by dynamic surface tension measurement, the release of active ingredient varied from less than 5 min to greater than 4 h through formulation modifications. Bioefficacy studies with the approved MMF G formulation against *Aedes albopictus* pupae gave > 90% mortality in 48 h, consistent with the mortality observed with liquid AGNIQUE® MMF product.

# 89 Mosquito control in backyard swimming pools in New Orleans, LA after Hurricane Katrina Claudia Riegel, criegel@cityofno.com, Mieu T. Nguyen, Cynthia Harrison, Gregory K. Thompson, and Stephen Sackett

No US city has ever been impacted by a disaster of this scale. All methods of mosquito control were implemented after the disaster, including the use of adulticides, larvicides, source reduction, and biological control agents. The City of New Orleans has located 5,921 swimming pools and more are found every day. In addition, there are ponds, hot tubs, fountains, and other vessels that hold water which are unattended. Many of these pools have become important sources of mosquitoes such as *Culex quinquefasciatus*, *Anopheles crucians*, and *Cx. coronator*. There has been an aggressive program implemented to locate unattended swimming pools, ponds, fountains and introduce mosquitofish (*Gambusia affinis*) for long-term control. Swimming pools were located by using satellite imagery, real estate listings, inspections, and calls from the public. Stagnant water in these pools and ponds was treated concurrently with an indigenous, commercially prepared, bacterium (*Bacillus sphaericus*) and 30 to 50 mosquitofish. As of October 15, 2007, only 702 pools have been filled in or removed and 2,154 pools were fully operational. Long-term monitoring and treatment of swimming pools will be needed in order to reduce the health risks to the residents of New Orleans.

# 90 Fish production in salt marshes with mosquito control ditches and those with open marsh water management areas

David. J. Tonjes, david.tonjes@stonybrook.edu, Kimberly Somers, Keith Brewer, and Gregory T. Greene

In conjunction with a project to revise and optimize a mosquito control program for Suffolk County (Long Island), NY, approximately 80 acres of salt marsh in the Wertheim National Wildlife Refuge were altered from mosquito control grid ditches to a channel/pond style open marsh water management (OMWM) source reduction setting. Four yr of 3 times a year monitoring (including sampling pre-alteration and post-alteration) was conducted at 40 stations (20 treatment sites and 20 control sites). It is commonly thought that mosquito control ditches may increase overall fish production in salt marshes because the ditches increase favorable edge habitats; the data from Wertheim quantify production for ditched marshes, but also show that the channel/pond OMWM marsh areas were even more productive.

# 91 Reducing salt marsh mosquito production with open marsh water management

David J. Tonjes, david.tonjes@stonybrook.edu, Dominick Ninivaggi, Thomas Iwanejko, and Mary Dempsey

In conjunction with a project to revise and optimize a mosquito control program for Suffolk County (Long Island), NY, approximately 80 acres of salt marsh in the Wertheim National Wildlife Refuge were altered from mosquito control grid ditches to a channel/pond style open marsh water management (OMWM) source reduction setting. Five yr of larval surveillance are reported here. The sampling was conducted in 2 ways: one following protocols established for the Region V US Fish and Wildlife Service assessment of OMWM, using a fixed network of 84 stations across the marsh, and a second where breeding locations were sought across the 160 acres of treatment and control areas. Both surveillance methods clearly show statistically significant reductions in mosquito breeding. The larval decreases resulted in reduced larvicide applications across the treatment areas, although some pesticide use was deemed necessary.

# 92 Introduction to Emergency Mosquito and Mosquito-borne Disease Control Symposium Nolan Newton, Nolan.Newton@ncmail.net

This symposium will explore the post-disaster control of mosquitoes and the diseases they transmit. Topics include: Emergency control of mosquitoes after forest fires; uses of the EMAC system; example of the EMAC system after KS floods; the ASTHO/CDC publication chapter on emergency control: NC's experiences with emergency control; and, predicting and controlling disease transmission to prevent emergency control needs.

### 93 The coming storm

Rosmarie Kelly, rmkelly@dhr.state.ga.us, and Trey English

GA is vulnerable to tornadoes, flooding, fires, and other natural and man-made disasters that cause severe disruption of essential human services and property damage to public roads, utilities, buildings, parks, and other facilities. Depending upon climate conditions, mosquito populations following water-related disasters can increase to a level where they become a public health risk, making the restoration of vital services to the citizens of the affected area both dangerous and difficult. To assist affected communities in disaster recovery efforts, the GA Department of Human Resources, Division of Public Health, Notifiable Disease Epidemiology Section (NDES) has acquired the resources and developed plans for mitigating the impact of post-disaster mosquito populations. A mobile laboratory trailer and personnel are available to support surveillance of vector and nuisance mosquito species in affected areas. This talk will provide information about post-disaster mosquito response, GA's mobile laboratory, and support resources available to GA communities and surroundings states that have mutual aid agreements with GA.

### 94 EMAC Attack: The Emergency Management Assistance Compact

Angela Copple and Frederick Frey, ffrey@mema.state.md.us

The Emergency Management Assistance Compact (EMAC) is a national mutual aid compact administered by the National Emergency Management Association (NEMA) that can be used by states to share resources during declared disasters. It facilitates deployment by settling, before an event, important issues such as licensing, liability, and reimbursement. Discussion includes the administration and organization of the compact. Also included are current items for process improvement. These include resource typing, standardizing mutual aid, and lessons learned from the effective EMAC deployments during the 2005 hurricanes season.

# 95 Rapid mosquito and mosquito-borne virus surveillance after floods in KS, July-August 2007 Bruce Harrison and Parker Whitt, pwhitt@triad.rr.com

In July and August 2007 two personnel from Public Health Pest Management Section, NCDENR were contracted and traveled to southeastern KS to conduct 10 nights of mosquito and mosquito-borne virus surveillance after severe floods in 4 counties. This trip was part of a FEMA Emergency Management Disaster Response. Three to 5 gravid and/or CDC miniature light traps were placed in each of these counties each night where flooding took place around homes and populated areas. Over 10,000 specimens of 29 species were collected, identified, and sorted to species. Four *Culex species* were targeted for pooling and shipped to CDC (Ft. Collins, CO) for virus testing. Over 4,000 specimens of *Culex* in 235 pools were tested, of which 30 pools were positive for West Nile virus.

### 96 Guidelines for emergency response in communities with limited resources

Daniel Sinclair, Chester Moore, Culex.tarsalis@comcast.net, Wayne Kramer, and Thomas Dunlop

The Association of State and Territorial Health Officials (ASTHO) is adding a chapter on emergency response to its publication, "Public Health Confronts the Mosquito: Developing Sustainable State and Local Mosquito Control Programs" (available at: http://www.astho.org/pubs/FinalReportPDF.pdf). Intended for wide distribution to local, state, and national stakeholders, the latest chapter explores issues in emergency management planning during disease outbreaks and natural disasters. The added chapter covers a number of issues and potential solutions, including the role and importance of effective surveillance, the proper timing of response activities, the role of risk assessment and communication to the public, the proactive involvement of all stakeholders, and the selection of appropriate and effective mosquito control measures. We focus on areas of particular interest to under-funded programs, such as resource sharing and access to the Emergency Management Assistance Compact (EMAC).

# 97 NC's experiences in post-hurricane mosquito control

Nolan Newton, Nolan, Newton@ncmail.net

When Hurricane Fran hit NC in 2006 our state emergency mosquito control plan that had been in place for about 20 yr, was put to a severe test. We modified the plan during post-hurricane evaluations and put the new plan into effect when Hurricane Floyd hit North Carolina in 1999, again doing post-response evaluations and modifications. After Hurricane Isabel hit in 2003, we once again evaluated our plan and made changes. The importance of planning, and planning for your plan to fail in some areas, will be discussed. Some of the problem areas within emergency response planning and the sometimes counter-intuitive ways to deal with these problems will be discussed. Strategies for dealing with emergency mosquito control issues require patience and a willingness to change plans quickly.

### 98 The effect of adulticiding on mosquito density in Chicago

John-Paul Mutebi, Mutebi\_JohnPaul@cdph.org

The city of Chicago used multiple ground ultra-low-volume (ULV) treatments of sumithrin (ANVIL 10+10) in areas with high West Nile virus (WNV) infection rates among wild *Culex* mosquitoes. The initial treatment reduced adult *Culex density* by 4.9 mosquitoes/trap/d and after the second consecutive treatment *Culex* density was reduced by 10.0 mosquitoes/trap/d. The density differences were statistically significantly different from the pre-treatment density levels, P<0.3 and P<0.01 respectively, and the *Culex* densities were reduced to very low levels. The treatments were discontinued for 7 d and the wild *Culex* density rebounded to pre-treatment levels. The third

treatment reduced *Culex* densities by 7.1 mosquitoes/trap/d (P<0.2), but the last treatment at the beginning of September had no effect on population density. Overall, multiple ground ULV treatments suppressed wild *Culex* density and this supports the recommendations by the CDC for controlling *Culex* mosquitoes especially *Culex* pipiens.

99 Knowledge and attitudes toward prevention and control of dengue in a slum area in Brasilia, Brasil
Ivenise Leal Braga, ivenise.braga@saude.gov.br, Ricardo Pimenta Faria, Iraci Crillanovick, Lígia Maria Paixão
Silva, Marisa Pacini Costa, and Ima Braga

A knowledge, attitude, and behavior survey was conducted among residents of a slum area in Brasília, Brasíl regarding dengue fever (DF) and dengue hemorrhagic fever (DHF). A random sample of 421 houses (2.1% of the area) was visited and a questionnaire was applied. About 94% of those interviewed had heard about dengue and 73.7% knew that the disease was transmitted by mosquito bites; 83.1% knew dengue symptoms and 14.4% informed us they had had dengue before; 47.5% knew the practices to avoid the mosquito found in containers and tires. Nearly 95% of slum-dwellers use barrels and drums for water storage, common sources of *Aedes aegypti*. The results show that adequate knowledge of dengue and prevention methods are found in close association with high *Ae. aegypti* domiciliary infestation rates. This suggests that traditional education strategies, although efficient in transmitting information, have failed to change the population's behavior. The study presents important data that appear to explain these attitudinal difficulties in avoiding infestations of household water storage containers because of sanitation problems in this community.

# 100 Serologic evidence for West Nile virus activity in two areas of Chiapas, México

Armando Ulloa, aulloa@insp.mx, José Domingo Méndez Sánchez, José Alfredo Juarez, Mauricio Casas Martinez, Juan Guillermo Bond Compean, Rogelio Danis Lozano, José Arturo Farfan Ale, Julián García Rejon, Luis Flores Flores, and Nicholas Komar

West Nile virus (WNV) is a member of the Japanese encephalitis virus complex within the genus *Flavivirus*, family Flaviviridae. In Chiapas, a serologic survey of 200 domestic animals conducted during 2001 detected a single animal (cow) with WNV-neutralizing antibody, a result considered inconclusive because of the complexities of determine the prevalence of WNV in several communities from the "Lacandon Forest" and the "Encrucijada," 2 natural protected areas in Chiapas. We sampled blood from 343 domestic animals, including 215 chickens, 50 turkeys, 44 cattle, 34 ducks, 4 horses, 2 geese, 2 pigs and tested them for WNV-specific antibodies using a blocking ELISA. Of 36 reactive sera, 19 samples were confirmed by plaque-rerduction neutralization test, including 8 chickens, 5 cattle, 4 turkeys, 1horse, and 1 goose. After combining results for all species tested, the serologic prevalence in Benemérito de las Américas, Roberto Barrios and Nuevo Orizaba from Lacandon Rain Forest was 19% (6/31), 7% (4/56) and 5% (2/42), respectively. Seroprevalence was lower (X2=5.6, P=0.03) on the Pacific coastal plain in the communities of Rio Arriba (6%, 6/94) and Zacapulco (1%, 1/120).

## 101 First West Nile virus isolations from mosquitoes in the Neotropics

Roberto Barrera, rbarrera@cdc.gov, Manuel Amador, Annette Diaz, Joshua Smith, Elizabeth Hunsperger, Jorge L Munoz-Jordan, Kovi Bessof, Manuela Beltran, Edgardo Vergne, Mark Verduin, and Wellington Sun

There is ample evidence of West Nile virus (WNV) circulation in the Caribbean and other tropical areas of Latin America. Virus identification in the region has been difficult because of an absence of patent epizootics or epidemics. WNV surveillance in North America has used dead birds, horses, sentinel chickens, and mosquitoes. We are not aware of surveillance programs in tropical America using sentinel chickens coupled with mosquito trapping to detect active WNV transmission and isolate the virus. We initiated a sentinel chicken WNV surveillance program in July 2006 in eastern Puerto Rico, targeting areas where WNV antibodies had previously been detected in a resident bird and 3 horses in 2004. Surveillance was conducted by taking blood samples every 2 wk from 60 non-immune chickens distributed across the 6 most representative terrestrial habitats. Blood specimens were tested by a chicken-specific IgM MAC-ELISA that would reflect a recent infection. The first positive chickens were detected in June 2007 in evergreen and mangrove forests. After this information was obtained, mosquito trapping was initiated around the sites where the cages were located and blood samples were taken weekly. Within 2 wk, WNV seroconversions were detected in all habitats. The WNV virus was isolated from *Culex nigripalpus* and *Cx. bahamensis* and also detected in *Cx. quinquefasciatus* by RT-PCR. This is the first report of WNV isolations from mosquitoes in the Neotropics. We recommend sentinel chicken surveilance coupled with mosquito trapping to detect recent or active transmission of WNV in the tropics.

# 102 Culex mosquito distribution in Tecamac, Mexico State, Mexico

Karla Balderrama-Martínez, kreepkar85@hotmail.com, Alvaro Diaz-Badillo, América Alejandra Padilla-Viveros, and Maria de Lourdes Munoz

West Nile Virus (WNV) was initially isolated in America from *Culex* species and birds in New York City area in 1999. Subsequently, the virus spread in the United States of America (USA) and many human cases have been reported. *Culex* mosquitoes are the most important vectors, although WNV has been isolated or detected in more than 20 mosquito species in USA. WNV was previously detected in equines and birds in Nuevo Leon, Tamaulipas, Coahuila and Yucatan states in Mexico. Because of the importance of this vector, the objective of the present research was to investigate the distribution of *Culex* species in Tecamac, Mexico State, Mexico, since the states of Mexico, Jalisco, Oaxaca, Michoacan, Tlaxcala, Saltillo, Nayarit and Morelos are potential states where WNV can

spread in Mexico. During May 2007, a total of 32 sites were sampled for mosquito eggs, larvae, and pupae. *Culex quinquefasciatus* was the dominant larval species collected. The likely importance of this species in the natural history of WNV in the northeastern USA prompted us to evaluate the oviposition sites of *Culex* species and associated species as potential future foci of WNV transmission in Mexico. We have demonstrated the presence of *Cx. quinquefasciatus* in the State of Mexico.

103 Preferred indoor resting sites of *Culex quinquefasciatus* Say in Monterrey, Nuevo Leon, Mexico Maricela Laguna-Aguilar, A. E. Flores, Gustavo Ponce-García, Ildefonso Fernandez-Salas, ifernand@fcb.uanl.mx

The study of adult *Culex quinquefasciatus* Say mosquitoes and the ambient dwelling factors explaining its presence are analyzed in this study. Description of this species' preferred resting was the goal of this work. Abundant *Cx. quinquefasciatus* populations are found in human settlements in Monterrey, northeastern Mexico. This fact represents a potential for West Nile virus (WNV) transmission, a disease with low endemicity to date but with the potential to cause large human outbreaks. Mosquito collections were made inside and outside houses using a backpack aspirator. Several variables were recorded concurrently, such as resting site, height above the floor, wall material and color, as well as blood digestion stages. The collected data were analyzed using the statistical program SPSS 10. Results showed a clear preference of this species to rest in bathrooms and bedrooms with smooth and rough wall surfaces. In addition, wall material was mostly dark colored cement. The higher presence of resting mosquitoes was recorded when the wall had less than 50 cm height. Most of resting sites had an average relative humidity of 50-60% and an average temperature of 30-35 °C. The distribution of females according to blood digestion status showed that gravid female *Cx. quinquefasciatus* preferred indoor resting sites. We concluded that indoor environments in Monterrey provided enough resources to allow this incriminated WNV vector species to potentially maintain human epidemics.

104 Host-feeding preference of Culex quinquefasciatus in Yucatan State, Mexico

Julian Garcia-Rejon, grejon@uady.mx, Jose Farfan-Ale, Maria Loroño-Pino, Wilberth Chi-Chim, Luis Flores-Flores, Elsy Rosado-Paredes, Carlos Baak-Baak, Bradley Blitvich, Jose Perez-Mutul, Victor Suarez-Solis, Ildefonso Fernández-Salas, and Barry J. Beaty

Studies were conducted to determine the host-feeding preference of *Culex quinquefasciatus* in urban areas of Merida, Yucatan State, Mexico. Mosquitoes were collected in the backyards of houses using wooden resting boxes. Collections were made 5 times per wk from January to December 2005. DNA was extracted from engorged females and tested by PCR using universal avian and mammalian-specific primers. DNA extracted from avian-derived blood was further analyzed by PCR using primers that differentiate among 3 avian orders: passeriformes, columbiformes and galliformes. PCR products obtained from mammalian-derived blood were subjected to restriction enzyme digestion to differentiate between human, dog, cat, pig, cow, and horse-derived blood meals. Overall, 82% of engorged mosquitoes had fed on birds and 18% had fed on mammals. The most common sources of avian-derived blood were galliformes and passeriformes. The most common sources of mammalian-derived blood were dogs and humans. The overall human blood index (HBI) was 6.7%. The overall forage ratio for humans was 0.1 indicating that humans are not a preferred host for *Cx. quinquefasciatus* in Merida.

105 Control of culicine mosquito larvae (Insecta: Diptera) using micronized suspensions of calcium hydroxide A. Martínez-Ibarra, A. Estrada Aguilar, R. M. Sánchez Manzano, A. D. Camacho, and B. Nogueda-Torres, bnogueda@yahoo.com

Laboratory bioassays and preliminary field tests were conducted to evaluate the larvicidal effect of micronized suspension calcium hydroxide (the main component of lime) and the median lethal dose ( $LD_{50}$ ) was determined. Fourth instar mosquito larvae were kept in a mixture of drinking water and original pond water from selected locations (Mexico City for *Culex* spp. and Ayala City, Morelos state for *Aedes* spp.). The control with Temephos® at 0.1 ppm (commercial concentration) caused 100% mortality of larvae 6 h after application and also killed pupae 28 h post-application. The  $LD_{50}$  for calcium hydroxide for *Culex* spp. larvae was 0.084% (w/v) 24 h after application, for *Aedes* spp. the  $CD_{50}$  for larvae was 0.035%. The effects of calcium hydroxide on pupae showed much variation; therefore, we do not consider this compound effective against pupae. Preliminary field testing conducted in ponds in Ayala City indicate that calcium hydroxide seems an effective larvicide at  $LD_{50}$ . Our results show the potential of micronized suspensions of calcium hydroxide as mosquito larvicide. It is worth mentioning that lime is cheap and the people in the rural areas of Mexico and other countries are familiar with the handling procedures for this compound.

Brain cell karyotype of *Culex quinquefasciatus* (Diptera: Culicidae) at three temperatures from Colombia Jesus Escovar, jeescobar@lalsalle.edu.co, Carolina García, Yesica Londoño, Ligia Moncada, and Estrella Cárdenas

The objective was to analyze the chromosomes of *Culex quinquefasciatus* at 3 temperatures. The adults were collected in the following places: El Muña (elevation 2,565m and 15 °C avg.), Fusagasugá (elevation 1,700m and 20 °C avg.) and Girardot (elevation 289m and 28°C avg.) in Cundinamarca Department, Colombia. Fourth instar larvae were obtained from first generation of the 3 colonies. The larvae were placed in colchicine; the heads were then placed in orcein stain on a microscope slides and the tissue was squashed. Forty slides were examined and mitotic chromosomes were photographed at 1,000X. The chromosomes were identified according to size and centromere position. The diploid number of *Cx. quinquefasciatus* was 2n=6 for the three populations. The length chromosomes were the following: the pair I shoved 2.82, the pair II 4.02 and the pair III 4.36 microns average at 15

°C; the pair I shoved 3.39, the pair II 4.57 and the pair III 5.05 microns average at 20 °C; and the pair I shoved 2.63, the pair II 3.32 and the pair III 3.85 microns average at 28 °C. The chromosomal measurements showed a greater size in the chromosomes of *Cx. quinquefasciatus* from Fusagasugá although the differences were not statistically significant.

# 107 Life cycle of Culex quinquefasciatus (Diptera: Culicidae) at different temperatures in Colombia Jesús Escovar, jeescobar@lalsalle.edu.co, Carolina García, Yesica Londoño, Ligia Moncada, and Estrella Cárdenas

The objective of this study was to determine the life cycle of *Culex quinquefasciatus* at 3 temperatures. The adults were collected in the following places: El Muña (elevation 2,565 m and 15 °C avg.), Fusagasugá (elevation 1,700 m and 20 °C avg.) and Girardot (elevation 289 m and 28 °C avg.) in Cundinamarca Department, Colombia. Laboratory colonies of this mosquito were maintained to 15 °C, 22 °C and 28 °C, according to origin. We observed that the life cycle of *Cx. quinquefasciatus* was 47.8 d average at 15 °C; while at 20 °C, it was 29.4 d average; and 35.9 d average at 28 °C. The life cycle of *Cx. quinquefasciatus* from El Muña (15° C avg.) was 38.9 d average at 22 °C and 32.4 d average at 28 °C; while mosquitoes from Fusagasugá (20 °C avg.) showed a life cycle 39.9 d average at 15 °C and 31.3 d average at 28 °C; and *Cx. quinquefasciatus* from Girardot (28 °C avg.) showed a life cycle 41.6 d average at 15 °C and 28 d average at 22 °C. The differences in life cycle among the 3 populations of *Cx. quinquefasciatus* were statistically significant. The results suggest a good capacity of adaptation by *Cx. quinquefasciatus* to environmental changes.

#### 108 Introduction to the Pesticides and the Environment

Rui-De Xue, xueamcd@bellsouth.net, Linda Wampler, and Jeanne Moeller

The purpose of this symposium is to provide an overview of the impact of mosquito control pesticide applications on the environment. Factors influencing pesticide efficacy in order to improve mosquito control effectiveness while protecting the environment will be discussed.

# 109 Climate change: Potential affect on pesticide application for vector control

Kenneth Linthicum, Kenneth, Linthicum@ars.usda.gov, and Assaf Anyamba

Global climate change has and will in the future contribute to the global burden of vector-borne disease by affecting the spatial and temporal distribution of disease. These changes in disease distributions are a direct result of altering the ecology of immature and adult habitats of insect vectors. The control of vector-borne disease is almost solely dependent upon vector control, which is conducted using the principles of integrated vector management (IVM). IVM is dependent upon accurate knowledge of the environmental and ecological conditions associated with the targeted species, and the appropriate use of control tools, including pesticides. Climate also directly affects pesticide applications including longevity, drift and deposition, and ultimately efficacy. We discuss how increases in temperature and changes in rainfall patterns can increase mosquito populations and vector capacity, and reduce efficacy of pesticide applications. Advanced understanding of climate change will permit the design of optimal pesticide application strategies to control vectors effectively.

# 110 Characterization of pesticide dispersion from aerial applications of mosquito adulticides: Meteorological effects and canopy penetration

Jane A.S. Barber, jasbarber@knology.net, Mike Greer, Mark Latham, and Gail Stout

Aerial adulticiding is a necessary tool which plays a vital role in the prevention of vector-borne diseases. There is, however, pressure upon the public health community to optimize applications both in terms of mosquito control and non-target health. We have, therefore, investigated the dispersion of permethrin at 2 application altitudes (30 and 45 m) over a range of typical atmospheric conditions. Chemical dispersion was physically characterized and biologically assayed as both flux and deposition. Wind speed fluctuations were measured via 4 separate sensor groupings: Three-dimensional sonic anemometers within (2 m) and at 2x canopy height (7 m); 2-dimensional anemometers within (2 m) and at canopy top (3 m); propeller anemometers at 2 m in the open and at application altitude via a kitoon\_system (30 and 45 m); and aircraft-mounted sensors at the delivery zone (30 and 45 m). Data is presented on the physical and biological characterization of spray dispersion. Overall, there was little difference in wind speed from one test to the next. There was however, a clear difference in atmospheric turbulence during treatment nights. The effect of stability on deposition and flux differed as altitude changed. When non-target mortality was high at 30 m application altitude the conditions were stable, and large volumes of pesticide descended via aircraft vortices and sedimentation. Overall, the 45 m application altitude did not produce excess non-target mortality regardless of stability at time of application. Neutral conditions with increased turbulence provided more operative control as effective flux.

### 111 Pesticide efficacy and mosquito behavioral impacts

Theeraphap Chareonviriyaphap, faasthc@ku.ac.th, John Grieco, Nicole Achee, Wannapa Suwondkerd, and Piti Monklangkool

Chemicals protect humans from the bite of insects using 3 different actions: Irritation after making contact, repelling prior to contact, or by killing the insects (toxicity). Most research has focused on the toxic function of chemicals, whereas comparatively few have concentrated on non-toxic chemical characteristics. In this study, we tested 3 actions of test chemicals on mosquito populations using both laboratory and field assay systems. Laboratory investigation was conducted using 2 systems: the free choice excito-repellency (ER) test box and the high throughput screening system (HITSS). Field studies were performed using experimental huts. Our findings indicate that test chemicals successfully repelled mosquitoes from treated surfaces at low concentrations, whereas higher doses provided a toxic action on mosquito populations. In particular, DDT demonstrated a unique property of "repellency," whereas synthetic pyrethroids (i.e., deltamethrin, permethrin, cyphenothrin, lambdacyhalothrin, alpha-cypermethrin, tetramethrin) primarily functioned as 'irritants'. The toxic action of chemicals at higher doses could stimulate selection for resistance in vector populations, whereas applying chemicals at minimal doses can help delay physiological resistance and possibly reduce the environmental risk.

# 112 Permethrin decomposition after 4-month storage in the spray truck tanks during mosquito-off season Rui-De Xue, xueamcd@bellsouth.net, Whitney Qualls, He Zhong, and Cate Brock

The concentrations of permethrin in the Aqua-Reslin® formulation stored in the insecticide mixers/pumps, 16 ULV spray truck tanks, and a stock container were sampled and analyzed by gas chromatography. The result showed that an average of 55.53% and 70.32% permethrin in Aqua-Reslin® were decomposed in the ULV spray truck tanks and in the mixers/pumps, respectively, during the 4 mo mosquito-off season. The degradation may directly result in economic loss. Also, mosquito control efficacy may be compromised if there is no appropriate concentration. Permethrin analytical method, economic cost, and degradation prevention methods have been discussed.

### 113 Sampling and analysis of barrier treatments

Brian Quinn, brian.quinn@ars.usda.gov, Ulrich Bernier, and Daniel Kline

Sampling vegetation correctly from barrier treatments is paramount to production of good chemical residue results. Representative samples from an entire area or subsection of a treated area provide a more accurate depiction of the chemical residues found in the vegetation, whereas grab samples will provide data from only a small section of a barrier treatment. Subsequently pairing these chemistry data with biological efficacy data of mosquito populations in barrier-treated areas creates a better understanding of the concentrations needed to provide protection for humans and other animals. Barrier studies in FL and AR were conducted using bifenthrin to determine efficacy and chemical residue concentrations. These studies will better define application techniques and chemical choices for barrier treatments, thereby providing tools and knowledge for military personnel and mosquito control districts to effectively control biting arthropods.

# 114 Isolation of mosquitocidal bacteria with improved efficacy from various mosquito habitats in FL Hyun-Woo Park, hyun-woo.park@famu.edu

Entomopathogenic bacteria are important in the control of insecticide-resistant populations of insect pests and vectors of human diseases. Toxins produced by these bacteria are often species-specific and do not contaminate the environment as the toxins have lesser residual efficacy and are generally safe for non-target organisms. Among these entomopathogenic bacteria, *Bacillus thuringiensis* subsp. *israelensis* (*Bti*) has been used most widely and extensively in many regions of the world to control mosquito and black fly larvae. Although no resistance to *Bti* has been reported in the field yet, laboratory selection of mosquitoes with *Bti* Cry proteins results in high levels of resistance. Furthermore, mosquito resistance to any of these proteins results in significant cross-resistance to the others. Mosquitocidal strains of *B. sphaericus* also have been used to control larvae. However, as this bacterium contains only a single binary toxin, high levels of resistance to *B. sphaericus* have already been reported in the field in several countries. Consequently, there is an urgent need to search for and isolate new indigenous and more effective mosquitocidal bacterial strains. To find highly mosquitocidal bacteria producing novel toxins, sediment samples from various mosquito habitats in FL have been screened and selective isolates have been characterized for their potential use for mosquito control.

### 115 Aspects of the IGR methoprene in the environment

Doug VanGundy, dvangundy@central.com

The Insect Growth Regulator (IGR) methoprene was first developed for mosquito control in 1975. The discovery of methoprene ushered in the biorational approach to insect control. IGR's offer alternatives to traditional chemistries by offering a softer approach to insect control. Methoprene degradation occurs primarily by 3 mechanisms: UV light, oxidation, and microbial degradation. Degradation occurs rapidly with exposure to these mechanisms (<24 h). Methoprene must be specially formulated to protect it from rapid degradation. These formulations protect and provide the residual needed to control the emergence of adult mosquitoes. The primary degradates for methoprene are 7-methoxycitronellic acid and 7-methoxy citronellal. Methoprene is applied at low use rates. Mosquitoes are very susceptible to methoprene; water concentrations in the low, single digit parts per billion range are sufficient to control adult emergence. Methoprene is of low toxicity to non-target organisms, mammalian LD<sub>50</sub> is > 34,560. The

susceptibility of mosquitoes works in conjunction with low use rates that are generally below the  $LD_{50}$  of non-target aquatic organisms. Throughout its 30 year history in mosquito control, methoprene has a struck a balance between the need to control mosquitoes and the need to protect sensitive aquatic habitats.

# 116 Time of host-seeking activity of *Culex tarsalis* and floodwater *Aedes*, northern CO, 2006-2007 Marvin S. Godsey, Jr., mjg9@cdc.gov, and Harry M. Savage

Knowledge of peak periods of host-seeking activity is critical for effective and economical control of disease vector and pest mosquitoes. In 2006, we undertook studies to delineate the time of maximum host-seeking of *Culex tarsalis*, a principal vector of West Nile virus (WNV) in northern CO, and of the most abundant pest species in the area, *Aedes vexans*, *Ae. melanimon*, and *Ae. dorsalis*. Four sites were selected to represent urban, suburban/rural, rural, and natural habitats. Two John Hock collection bottle rotator traps programmed to collect at 2-h intervals from 1730-0730 h were placed at each site. A total of 74,464 mosquitoes were collected, of which 56,359 were *Ae. vexans*, 9,815 were *Ae. melanimon*, 3,801 were *Cx. tarsalis*, and 2,163 were *Ae. dorsalis*. Highest numbers of *Cx. tarsalis* (2,149) were collected at the natural site, followed by the rural site (888), suburban/rural site (610), and urban site (154). Preliminary analysis suggests that the peak time of host-seeking by *Cx. tarsalis* occurs during the 2130-2330 h time period, and that numbers taper off throughout the night. Data will also be presented on seasonal and site-specific differences in abundance of the four species.

# 117 A quantitative in vitro assay for chemical mosquito-deterrent activity without human blood cells Jerome A. Klun, Jerome.Klun@ARS.USDA.GOV, Matthew Kramer, Aijun Zhang, Shifa Wang, and Mustapha Debboun

We report that an aqueous solution containing 10-3 M adenosine triphosphate (ATP) and citrate-phosphate-dextrose-adenine (CPDA-1) can effectively replace transfusable human red blood cells in an in vitro K&D bioassay system for evaluating repellents, using either *Aedes aegypti* or *Anopheles stephensi* mosquitoes. These 2 species fed with similar avidity through collagen membrane covering aqueous 10-3 M ATP plus CPDA-1 or red blood cells in CPDA-1 supplemented with ATP. In a second experiment, we evaluated the feeding-deterrent activity of DEET and a newly discovered natural product chemical, (-)-isolongifolenone, against these 2 species. We found that the feeding-deterrent efficacy of the 2 chemicals was similar whether the feeding stimulant was red blood cells supplemented with ATP or ATP alone with CPDA-1. Since the use of human red blood cells in bioassays raises important health and logistic issues, aqueous ATP with CPDA-1 is a reasonable alternative to human blood cells for routine in vitro chemical screening.

# 118 Attractiveness of several octenol analogs and blends to Aedes albopictus and Culex quinquefasciatus James E. Cilek, cilek\_j@popmail.firn.edu, and C.O. Ikediobi

Several alkenol analogs of octenol were synthesized and evaluated against laboratory-reared *Aedes albopictus* and *Culex quinquefasciatus* in large walk-in screened cages. Some of the analogs were combined with several substances previously known as mosquito attractants to humans and animals. Results of these trials will be presented.

# 119 Anopheles darlingi - Amazon region main malaria vector - seasonality alterations from global climatic changes

Wanderli Pedro Tadei, tadei@inpa.gov.br, Iléa Brandão Rodrigues, Rosemary Costa Pinto, Joselita Maria Mendes dos Santos, Miriam Silva Rafael, Carlos Praia Lima, Antônio Evandro Melo Oliveira, Norbert Becker, and Thania Virgínia Guaycurus

Anopheles darlingi, the main seasonal malaria vector in Amazon, correlates to the flood pulse. During the flood pulse's receding water stage, the riverbank mosquito oviposition sites disappear, there is a reduction on population density, and the reproductive sites stay restricted to non-flooded land. There is correlation between malaria occurrence and An. darlingi seasonality. The effective measures for malaria control show a close relationship to entomological knowledge since it constitutes the primary information regarding this vector. Monitoring data on the Coari/Manaus gas-pipeline construction area, in the State of Amazonas, showed modifications on the species' seasonality, reflecting the responses to the climatic modifications that have been occurring in the Amazonian region. There was an unusually prolonged low water period in 2005; alterations in the normal flood pulse rhythm in late 2006 early 2007, provoked a heavy increase in An. darlingi density, in February, when this usually takes place between May and June. Anopheles darlingi collection indices ranging from 15 to 30 individuals in other months increased to 1,029 and 1,599 in February and March, respectively. Consequently, malaria incidence increased from February to April 2007 in the townships influenced by black-water rivers and lakes. Financial support was provided by PETROBRAS/FAPEAM/CTPETRO/PIATAM.

# 120 Generating CO<sub>2</sub> for mosquito monitoring: A comparison of mechanical vs. programmable CO<sub>2</sub> generators Reginald Coler, rcoler@iscatech.com, and Agenor Mafra

The prototype device developed can produce and deliver  $CO_2$  to a monitoring trap at any pressure and rate. This system produces  $CO_2$  gas (via a chemical reaction) only as needed and maintains the remainder of the  $CO_2$  in its more stable chemical form. The  $CO_2$  Generator, in its simplest form, does not require electricity and will release gas at a constant rate until the chemical reagents are spent. The electrical version of this unit is capable of releasing

 $\mathrm{CO}_2$  at specified intervals (time of day and the day of the week). This more sophisticated electronic version not only further conserves the unit's chemical agents (by releasing on specific days of the week and within a specified interval of time), but also allows the operator to pulse the output—effectively mimicking the  $\mathrm{CO}_2$  pattern of a breathing animal. Locating sources of dry ice and compressed gas often presents a logistical problem for mosquito monitoring personnel, especially in remote areas or military settings. In contrast, ISCA's  $\mathrm{CO}_2$  Generator produces pure  $\mathrm{CO}_2$ , using chemical reagents that are safe, easy-to-use, and, most importantly, can be shipped directly to any address with a guarantee for extended shelf-life.

# Mosquito diversity compared to USGS continuously recorded river levels in a Neuse River NC floodplain Alice L. Anderson, andersonal@ecu.edu, and Joseph Sutton

Twice weekly samples using CDC light traps were collected over 1 year starting in the fall 2006 in a Neuse River watershed tributary floodplain. The USGS in NC has monitored water levels in the 2 main watersheds for many years and is now planning to discontinue this real-time data flow. Interesting data on drought conditions such as those developed over the 2007 season will be lost. In this project, we compare the fall 2006 seasonal diversity in this habitat to the real-time fluctuation of river water data collected by the USGS in NC during the same time period.

# 122 Surveillance of *Culex pipiens* L. populations for resistance to d-phenothrin in Prince William County, VA Filipinas M. Caliboso, FCaliboso@pwcgov.org, Karrie L. Cox, Karen A. Walker, and Benjamin F. McLaurin, Jr.

The monitoring of insecticide susceptibility in mosquitoes is a vital component of any pest management program for early detection of build-up of resistance to insecticides in use. The results of a surveillance program to detect the development of resistance to d-phenothrin of *Culex pipiens* populations within Prince William County, VA, are reported. Adopting the CDC bottle bioassay procedure, baseline susceptibility was established using a wild strain of the test species. Time-mortality response was determined in 9 field-collected populations and compared with the reference population. No resistance was detected in all strains tested.

#### 123 Withdrawn

### 124 Larviciding vs. adulticiding: Why larviciding may be a better choice

Vince Sanchez, vsanchez5195@sbcglobal.net

Larviciding vs. adulticiding: Materials, methods, and costs of mosquito control will be discussed based on a Senior Mosquito Control Technician's experience in a salt marsh area of CA's Central Coast. The biological and financial advantages of using larvicides to kill mosquitoes before they become flying adults will be emphasized.

# 125 Over 50 years of observations on the efficacy of ground adulticiding

John Beidler, dcarlson1@hotmail.com

This presentation will summarize my over 50 years of experience with ground adulticiding both from an operational and research perspective. While ground adulticiding is what most people first associate with mosquito control, it is only one part of an integrated program approach. My presentation will discuss effective ground adulticiding dosage rates, comparisons with aerial adulticiding, and some of the recent technological advances in this field.

# 126 Ground ULV as a tool for adult mosquito control

Kenneth Townzen, ktownzen@sbcglobal.net, and Dan Ariaz

Ground ULV for control of adult mosquitoes is an important component in a comprehensive program. When disease is present or control of larval sources are cost prohibitive or impossible to reach, adult mosquito control is necessary. To be successful, ground ULV tactics include equipment that produces the proper drop size, a viable product applied properly, and weather conditions that allow the insecticide droplets to disperse throughout the target area. Periodic evaluation applications will support confidence in this important mosquito control tool.

# 127 Basic considerations concerning the efficacy of aerial adulticiding operations

Mark Latham, manateemcd@aol.com

Aerial adulticiding is an important tool to many mosquito control programs. It can be conducted using aircraft owned and operated by the program, or by private contractors. The advantages of aerial adulticiding over the use of truck adulticiding include the ability to treat large areas in a short period of time and the ability to treat areas lacking a good road network. Disadvantages include higher cost, greater reliance on appropriate meteorology to get the spray to the target zone, greater risk of spray drift out of the target zone, and safety concerns resulting in higher than ideal spray altitudes. Aerial adulticiding is not appropriate for all areas or all target mosquito species and should only be used after careful consideration.

# 128 Observations on the bionomics and ecology of *Anopheles (Nyssorhynchus) marajoara* (Diptera: Culicidae) in Venezuela

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Anopheles (Nyssorhynchus) marajoara Galvão and Damasceno has a wide geographic distribution in Venezuela. To contribute to the knowledge on the bionomics and ecology of this malaria vector, studies were conducted to identify

and characterize larval habitats. Collections of adult females from light traps and on human baits were carried out in different localities in the states of Apure, Amazonas, Barinas, Bolívar, Delta Amacuro and Guárico. This species was found at elevations between 35 and 220 m, in different ecosystems such as rain forests, gallery forests, savannahs, secondary vegetation and rice, plantain, and cotton plantations. Field-collected females laid a mean of 78 eggs; the development from egg to adult takes 10 d at 28° C. Larvae were collected mainly in stream margins and semi-permanent lagoons associated with *An. nuneztovari, An. argyritarsis, An. darlingi, An. braziliensis, An. oswaldoi, An. strodei, An. costai,* and *An. punctimacula. Anopheles marajoara* is more abundant during the rainy season (May-December) with a peak in August. Regarding the diel biting pattern, this species bites throughout the night indoors and outdoors, although significantly more mosquitoes were collected outdoors (>70%) and 75-83% mosquitoes were caught before midnight. The higher adult mosquito densities were found in Guárico State in sites close to rice fields.

# 129 Genetic structure of Anopheles (Nyssorhynchus) marajoara (Diptera: Culicidae) in Colombia Helena Luisa Brochero, embrochero@unal.edu.co, Cong Li, Richard Wilkerson, Jan Conn, and Manuel RuízGarcía

Five Anopheles marajoara populations, representing diverse ecological conditions, were sampled throughout Colombia and analyzed using 9 variable DNA microsatellite loci. The overall genetic diversity (H = 0.58) was lower than that determined for some Brazilian populations using the same markers. The Caquetá population in Colombia had the lowest gene diversity (H = 0.48) and it was the only population at Hardy-Weinberg equilibrium. Hardy-Weinberg disequilibrium in the remaining 4 populations was probably due to the Wahlund effect. The assignment analyses showed 2 incompletely isolated gene pools separated by the Eastern Andean cordillera. However, other possible geographical barriers (rivers and mountains) did not play any role in the moderate genetic heterogeneity found among the populations (FST = 0.069). These results are discussed in relation to An. marajoara's possible role as a malaria vector.

# 130 Identification and taxonomic position of *Anopheles nuneztovari* (Diptera: Culicidae) from the Brasilian Amazon

Daniela Calado, danielacalado@hotmail.com, Cecilia Luisa Simoes dos Santos, Raimundo Nonato P. Souto, Allan Kardec R. Galardo, and Maria Anice Mureb Sallum

Anopheles nuneztovari has extensive distribution and occurs in northern South America and eastern Panama. Cytotypes A, B and C are recognized; only cytotype A was reported from Brasil. Considering that this taxon may comprise a species complex and cytotype A represent a distinct species from Colombian (C) and Venezuelan (B, C), our goal is to clarify the taxonomic status of An. nuneztovari A. We used 20 individuals identified by male genitalia. ITS2 and COI sequences were obtained from samples collected in: Itacoatiara and Parintins (Amazonas), Prainha, Almeirim and Óbidos (Pará) and Macapá (Amapá). Only 2 ITS2 amplicons were direct sequenced and showed 100% similarity. Due to intragenomic variations, we cloned 18 samples. Intraindividual ITS2 variability was observed in microsatellite regions (GA). ITS2 sequences could not separate cytotype A from B and C. Variations used to separate cytotypes B/C from A were observed in some clones from cytotype A. Our results suggest that ITS2 spacer is not suitable to unambigously distinguish any cytotypes. COI revealed that Brasilian samples are distinct from those from Venezuela and Colombia. We identified An. dunhami using molecular and male genitalia characters, 1 specimen was recognized using ITS2 sequence only. Anopheles dunhami is first reported in Parintins.

# 131 Efficacy and persistence of methoprene on *Anopheles albimanus* (Wiedemann) pupae Jesus Berti Moser, jbertimoser@yahoo.com, and Edith Navarro

Anopheles albimanus Wiedemann has been considered under natural conditions as human malaria's principal vector in Central America. Methoprene is a synthetic juvenile hormone and is used in the larval control of vector mosquitoes. This research contemplates the realization of a series of experiments under laboratory conditions with a granular formulation of methoprene (Altosid-G<sup>®</sup> 1.5%) and An. albimanus larvae, to calculate the lethal concentrations of the product, as well to evaluate its efficacy and persistence. The aim of this study was to evaluate the efficacy and persistence of lethal effect of methoprene on An. albimanus pupae and to calculate the lethal concentrations (CL50 CL95 CL98) of the product under laboratory conditions. The product's persistence over An. albimanus colonized larvae was evaluated using the lethal concentration 95 (CL95) of 0.1515 ppm, at 0, 7, 15, 30 and 60 d post-treatment. According to the results obtained at 0 and 7 d post-treatment, the product's efficacy was very satisfactory, with 95% and 75% mortality. But the percentage of dead pupae in treated water diminished at 15, 30 and 60 d post-treatment. In conclusion, this granular formulation has a short residual activity on An. albimanus pupae and the permanence of the product in treated water (15, 30 and 60 d) negatively affected its effectiveness.

# 132 Isolation and selection of stocks of fungi for *Anopheles albimanus* mosquito control M.G Vázquez-Martínez, mguadalu@insp.mx, O.R. Gálvez-Coutiño, and A. Rodríguez-Meneses

Recently, 2 studies demonstrated that entomopathogenic fungi can kill adult anopheline mosquitoes and open new opportunities in the fight against malaria. The present study was carried out to isolate, identify and select entomopathogenic stocks from native fungi associated with *Anopheles albimanus*. The isolations were made from water, ground and larval samples collected in mosquito oviposition sites in the southeastern coastal plane of Chiapas, Mexico. A total of 29 stocks were isolated; the most frequent were: *Aspergillus niger* (21.08%),

Penicillium spp. (17.69%), Gliocladium spp. (8.62%), Aspergillus terreus (8.31%), Geotrichum spp. and Rhizopus spp. (5.38%). The native stocks of Gliocladium spp., in addition to Beauveria bassiana and Metarhizium anisopliae, were evaluated for pathogenicity on An. albimanus 3<sup>rd</sup> instar larvae. Stocks of M. anisopliae showed the highest pathogenicity, causing 100% mortality during the first 24 h of exposure, to a concentration of 2.58x10<sup>7</sup> conidias/ml. Gliocladium spp. and B. bassiana showed 58.7% and 33.3% mortalities, respectively until 7 d post-inoculation. Stocks of M. anisopliae and Gliocladium spp. will be reactivated and re-evaluated on mosquitoes to determine lethal concentration (LC<sub>50</sub> and LC<sub>90</sub>) since they are good candidates for biological control of mosquitoes.

# 133 Quantification of *Bacillus sphaericus* produced by single larvae of *Anopheles marajoara* exposed to Vectolex WDG

Darjaniva Molina de Fernandez, darja2410@hotmail.com, and Luisa Figueroa Acosta

Anopheles marajoara larvae killed by application of Bacillus sphaericus (Vectolex® WDG 0.5; 1 and 1.5 Kg/Ha, Serotype H5a5b, strain 2362, 51.2%, 650 ITU/mg) were washed with a germicidal product to eliminate any traces of B. sphaericus on the external surface of the body. The larvae were placed individually in Petri dishes containing 20 ml of distilled water, replicated 10 times for concentration. In the first 6 h and 12 h, samples were taken from 10 microliters of each replication, which were inoculated in Lennox LB medium agar, incubated at 37° C for 12 h. Bacterial growth was observed with an uncountable number of colonies; subsequently at 18 h samples of dilutions of each medium 1x10-2, 1x10-4 and 1x10-6 were inoculated. We found that for the first 2 dilutions the number of colony forming units (cfu), was not quantifiable, while for dilution 1x10-6 showed bacterial growth with an average of 52 colonies per plate, which translates into 5,200 per ml. It was inferred that a board of 20 ml liquid medium, a single larva until 104,000 cfu, and one L 5.2 million (cfu). Sporule and non-sporule forms in the colonies isolated by smear were observed. This confirms the reproduction of B. sphaericus in an aquatic environment.

# High altitude malaria: Integral evaluation of malaria in a mountain ecosystem in Bolivia Marilyn Aparicio Efen, marilyneffen@yahoo.com.ar, James Aparicio, Jaime Rodriguez, Felix Trujillo, and Rimort Chavez

Climate change affects human health in both direct and indirect ways. One of the indirect effects is the increased incidence of vector-borne diseases in regions where those diseases have not been historically present. This happened in the Andean region of Bolivia near Lake Titicaca at an elevation of about 3,815 m in 1998 when there were malaria outbreaks in 8 communities and some cases were reported annually through 2006. This study carried out a climate change vulnerability and variability assessment, using a epidemiological integral assessment with a comprehensive ecosystem evaluation including the evaluation of biodiversity changes and vectorial habitat in the study area, climate analyses, clinical and laboratory tests, surveys (malaria symptomatology, homemade treatments, and ancestral knowledge related to climate and diseases), and using GIS and LANDSAT satellite images. The local malaria parasite (*Plasmodium vivax*), is the first noted at this elevation (2,615 to 3,590 m) related to climate change (0.7° to 1 °C of increments). These changes are producing new ecological conditions for vector presence (*Anopheles pseudopunctipennis*). Furthermore, the strong 1997-1998 ENSO event produced maximum historic temperatures and pluvial precipitation amounts in the study area. The study confirmed the local malaria outbreak of 1998 and unstable endemic presence of the disease in the region.

# 135 Elevated oxidase and esterase levels associated with DDT and lambdacyhalothrin cross-resistance in Anopheles darlingi from the Medio Atrato Region of Colombia

Idalyd Fonseca González, idalydf@guajiros.udea.edu.co, José D. Palacios, Silvia Díaz, and Martha L. Quiñones P

Currently, indoor residual spraying (IRS) and insecticide treated nets (ITN) are the mainstay in malaria prevention. These strategies use pyrethroid insecticides. In the Medio Atrato region of Colombia, IRS using lambdacyhalothrin is the main control measure. Earlier studies reported resistance to DDT in *Anopheles darlingi* from this site although the insecticide mechanism associated was not investigated. To evaluate the current susceptibility status and identify the biochemical mechanism involved in this population, we carried out WHO and CDC bottle bioassays and biochemical assays. The insecticides evaluated were lambdacyhalothrin, deltamethrin, permethrin, malathion, fenitrothion and DDT. Both methodologies detected high resistance to lambdacyhalothrin and DDT with mortality levels of 65% to 75%, respectively. Biochemical assays showed a significant elevation of oxidase and esterase activity compared with a susceptible population. The continued use of lambdacyhalothrin for the control of *An. darlingi* in this locality has resulted in resistance to this insecticide. In addition, DDT resistance is still present in this population; even though this insecticide has not been used in Colombia since 1992. These results suggest that metabolic detoxification by oxidases and esterases could be involved in cross-resistance between these two insecticides and demonstrates the urgent need to develop local insecticide resistance surveillance programs in malaria vector mosquitoes.

# 136 Insecticide resistance evaluation of the main malaria vectors in Colombia

Martha L. Quiñones, mlquinonesp@unal.edu.co, Idalyd Fonseca, Rocio Cardenas, Helena Luisa Brochero, Liliana Santacoloma, Myriam Salazar, Janet McAllister, and Clara Ocampo

After the ban on DDT usage and the decentralization of the malaria program in Colombia, different pyrethroid and organophosphate insecticides have been used for malaria control. Few local studies have been conducted of the insecticide susceptibility of the main malaria vectors. The purpose of this work was to update the susceptibility status of the three main vectors to these insecticides and to implement a national surveillance network of insecticide

resistance. Three research institutes and entomology teams of 12 states were involved in the project. Susceptibility tests were carried out using both the WHO and CDC methodologies. The insecticides evaluated were deltamethrin, lambdacyhalothrin, permethrin, cyfluthrin, DDT, fenitrothion, malathion, propoxur, bendiocarb, methyl-pirimiphos, and etophenprox. Out of six populations of *Anopheles albimanus* evaluated, 1 showed results compatible with resistance to lambdacyhalothrin. *Anopheles darlingi* showed resistance to DDT and lambdacyhalothrin in 1 population out of 9 evaluated, and *An. nuneztovari* showed resistance to deltamethrin, lambdacyhalothrin, DDT and fenitrothion in 2 populations out of 4 evaluated. The resistant populations coincided with the regions with higher annual parasite rates and, therefore, extensive use of insecticides for malaria control. Local entomology teams were trained to continue the surveillance and a national network for insecticide resistance of malaria vectors in Colombia was initiated. Financial support was provided by COLCIENCIAS.

# 137 Laundering effects on bite protection and permethrin content of Marine Corps Combat Utility Uniforms (MCCUUs)

Ulrich Bernier, uli.bernier@ars.usda.gov, Melynda Perry, Betsy Moore, Amy Johnson, Brian Quinn, and Gary Clark

The United States Marine Corps (USMC) has recently transitioned to factory production level permethrin treatment of their Marine Corps Combat Utility Uniforms (MCCUUs). As part of the qualification process for companies to supply the USMC with uniforms, these vendors must submit uniform specimens for validation. Validation requires that specimens meet both chemical and biological criteria. EPA label requirements stipulate that uniforms be treated with permethrin at an amount that adds 0.52% that of the total garment by weight. Therefore, USMC specifications require application of 0.125 mg/cm2 permethrin for blouses and 0.141 mg/cm2 level for trousers, not including a 10% tolerance of deviation from this value. Uniforms must also meet biological efficacy requirements of 96% protection at the unwashed and 20x laundered levels and 90% protection at the 50x laundered level. This presentation will present results from the chemical and biological analysis of the 4 suppliers of MCCUUs, examine the correlation between residual permethrin on the uniforms and the protection form biting, and explain why earlier permethrin treated MCCUUs exhibited rapid loss of bite protection after only a few launderings.

# Novel botanical-based insect pesticides and their environmental fates

Junwei Zhu, jwzhu@iastate.edu

The current presentation will discuss the recent advancement of the development of botanical-based insecticides and their impacts on environments. It will focus on the research conducted on developing new mosquito, cockroach, and fly repellents, as well as, larvicides and their potential uses on insect pests. The use of these newly developed pesticides will be evaluated for their environmental fates as well.

# 139 Solar exposure time impact on the efficacy of charcoal sustained s-methoprene against mosquito larvae Whitney Qualls, quallsamcd@bellsouth.net, Rui-De Xue, and Arshad Ali

The efficacy of charcoal sustained s-methoprene was evaluated to determine the effects of solar exposure on the mortality of 3<sup>rd</sup> instar *Culex quinquefasciatus* and *Aedes aegypti* larvae at 6-months, 1-yr, and 1.5-yr post-exposure.

#### 140 Scientific resolution of butterfly – mosquito control conflicts

Lawrence Hribar, lhribar@kevsmosquito.org

A cooperative research project was developed to investigate the effects of mosquito control operations on nontarget butterflies.

### 141 Mosquito adulticides' impacts on non-target organisms

He Zhong, zhongh@knology.net, and Cate Brock

Insects are the most diverse group of organisms on earth with an estimated 30 million species (Erwin 1982) of which only 1.4 – 1.8 million have been classified (Stork 1988). Mosquitoes comprise a very small fraction of the total insect species, with only 79 species identified in FL (Smith et al. 2006). Yet this small group of insects is capable of producing billions of individuals that could represent a significant public health threat and bring about substantial community discomfort. Mosquito adulticides are insecticides that are registered (or permitted) for use to control adult mosquitoes either by aerial or ground spraying. Current mosquito adulticides, such as naled, permethrin, and malathion are broad-spectrum insecticides that may affect non-target organisms directly exposed to them. If this exposure reaches the lethal limit for a specific non-target, it will die quietly without being noticed. For many years, some degree of non-target mortality linked to an adulticiding operation was accepted as a 'casualty of war'. But to what degree can we afford to wage this war at the adverse cost of non-targets? At what point do these casualties begin to compromise the natural balance of ecosystems? Adulticiding is necessary and beneficial – when properly applied with an environmental conscience. The authors encourage the thoughtful and responsible use of mosquito adulticides. More specifically, we support a balanced approach to mosquito control – that is, applying adulticides in a way that optimizes control efficacy and minimizes insecticidal impacts on non-targets.

### 142 Mosquito larvicides' impacts on non-target organisms Sharon Lawler, splawler@ucdavis.edu

A suite of options is available to control larval mosquitoes in the USA and abroad. Control materials vary in developmental stages affected, duration of activity, and non-target effects. Our lab has investigated non-target effects and efficacy of lambda-cyhalothrin (used internationally but not in the USA), temephos, Golden Bear Oil (GB1111), methoprene, and *Bti*. Materials were usually effective but various environmental or developmental issues can cause treatment failures for some materials. Non-target effects of lambda-cyhalothrin and GB1111 showed their potential to disrupt biological control. Nevertheless, GB1111 is useful because of its activity against pupae. Lambda-cyhalothrin and some methoprene formulations may persist in the environment causing potential for resistance. As in most studies, *Bti* was found to be safe for non-target species tested. The costs and benefits of these and other larval treatment options will be discussed.

# 143 Haemaphysalis phasiana (Acari: Ixodidae) in Korea: Distribution and habitat descriptions William J. Sames, mosquitodoctor@yahoo.com, Heung Chul Kim, and Terry A. Klein

In April 2007, tick surveys were conducted in Jeju Province and along the southern coast of Gyeongsangnam and Jeollanam Provinces, Republic of Korea, to assess the tick species composition and distribution, and to study habitat occurrence. Fifty-eight sites were sampled on Jeju and 19 and 25 sites were sampled in Gyeongsangnam and Jeollanam, respectively. Sampling was primarily conducted using tick drags through various vegetative habitats. Habitat descriptions were recorded for each 10 m drag and analyzed to determine habitat parameters for each collected species. During this survey, *Haemaphysalis phasiana* Saito, Hoogstraal, Wassef was collected at 10 sites (138 nymphs) in Jeju Province and at 6 sites (70 nymphs and 2 females) in Jeollanam Province. No *H. phasiana* were collected at the sites in Gyeongsangnam Province. Detailed habitat descriptions were acquired for this species in southern Korea.

# Reemergence, persistence, and surveillance of *vivax* malaria and its vectors in the Republic of Korea Terry A. Klein, terry.klein@kor.amedd.army.mil, Heung-Chul Kim, Leopoldo M. Rueda, Won-Ja Lee, Richard C. Wilkerson, Sung-Tae Chong, Jason G. Pike, and Robert G. Moore

Plasmodium vivax was endemic on the Korean Peninsula for many centuries until the late 1970s. It reemerged in 1993 and increased exponentially among ROK military and civilians through 1998. With the implementation of a chemoprophylaxis program by the ROK military in 1997, malaria stabilized and then decreased through 2004 as the use of chemoprophylaxis increased. Malaria cases increased in 2005-2006, but stabilized in 2007, most likely as a result of increased use of chemoprophylaxis. Taxonomic studies identified 2 new species belonging to the Anopheles sinensis complex in 2005. Extensive larval and adult surveillance identified larval habitats and geographical distribution of members of members of the An. sinensis complex. Anopheles sinensis sensu stricto was the most commonly collected anopheline throughout the ROK, followed by An. kleini and An. pullus. Geographical distribution models based on larval and adult surveillance indicate that An. kleini and An. pullus are relatively widespread throughout Korea. Population densities of An. kleini and An. pullus are highest in malaria high-risk areas. Data suggests that An. pullus and An. kleini are the primary vectors of P. vivax malaria in malaria high-risk areas near the DMZ while An. sinensis s.s. is a secondary vector.

# 145 A multispecies approach to predicting the geographic distribution of potential malaria vectors and malaria risk in the Republic of Korea

Desmond H. Foley, foleydes@si.edu, Terry A. Klein, Heung Chul Kim, Richard C. Wilkerson, and Leopoldo M. Rueda

Detailed knowledge of the geographical distribution of the total mosquito fauna known or suspected of being disease vectors for a given region would assist efforts to understand the epidemiology of vector-borne disease, the distribution of disease risk, and the extent and type of vector control required for that region. The anopheline fauna of the Republic of Korea comprises 8 species: *Anopheles belenrae* Rueda, *An. kleini* Rueda, *An. koreicus* Yamada & Watanabe, *An. lesteri* Baisas & Hu (=An. anthropophagus), *An. lindesayi japonicus* Yamada, *An. pullus* M. Yamada (=An. yatsushiroensis), *An. sinensis* sensu stricto (s.s.) Weidemann, and *An. sineroides* Yamada. We used adult and larval mosquito collection data from sites throughout the ROK and modeled the distribution of all 8 anopheline species using the programs Desktop Garp and MaxEnt. The resulting models of potential species distributions were compared to models of malaria suitability to better understand the distribution of malaria risk. A multispecies approach that matches predicted mosquito distribution to foci of malaria transmission could help incriminate malaria vector species in cases where the vectorial importance and distribution of individual species is poorly known.

# 146 The chikungunya outbreak in Emilia-Romagna Region, Italy, in 2007

R. Bellini, rbellini@caa.it, C. Venturelli, A. C. Finarelli, R. Angelini, P. Angelini, M. Dottori, and P. Macini

Chikungunya, an arbovirus mainly transmitted by *Aedes* mosquitoes, is endemic in some areas of Africa and Asia. In August 2007, an outbreak occurred, for the first time in Europe, in the Emilia-Romagna region (northern Italy). Evidence suggests that the index case arrived from India on June 15. On August 15, the local public health unit became aware of a possible epidemic going on. Entomological investigations showed haematophagous species present in the area were *Aedes albopictus*, *Culex pipiens*, and *Ae. caspius*. The chikungunya virus has been detected

and isolated from both the *Ae. albopictus* pools collected, while all the other pool results were negative. As of September 26, 306 suspected cases in 5 different foci were notified, of which 142 were confirmed and 49 tested negative. An emergency vector control program including adulticiding, larviciding, and source removal was conducted in public and private areas, in a radius of 100 m around each suspected case residence, and in a radius of 300 m around the foci (clusters of 2 or more cases). Blood and organ donations have been suspended in the affected municipalities. *Aedes albopictus* is widely present in Italy, and there is a risk of further outbreaks. Surveillance should be strengthened nationwide in order to promptly detect suspected cases and to implement efficacy control measures.

# 147 Laboratory and field evaluation of *Bacillus thuringiensis* var *israelensis* WDG against *Aedes albopictus* larvae in Lahore, Pakistan

Nusrat Jahan, dr.nusratjahan@gcu.edu.pk, and Saima Hanif

Laboratory and field evaluation of *Bacillus thuringiensis* var. *israelensis* Vectobac <sup>®</sup> WDG (Water dispersible granules) 3000 ITU/mg was carried out against *Aedes albopictus* (early 4<sup>th</sup> instar) larvae from Lahore, Pakistan. The primary objective was to determine the minimum effective dosage of *Bti* WDG for larval mortality and pupae reduction of *Ae. albopictus* in laboratory and in the field. Laboratory bioassays were carried out using 7 different concentrations (100, 10, 0.1, 0.01, 0.001, 0.0001 ppm) and revealed maximum larval mortality (100%) against high concentrations (100, 10, 1, 0.1 ppm) and minimum mortality (56, 44, and 20%) against low concentrations (0.01, 0.001, 0.0001 ppm). Probit-regression analysis indicated  $LC_{50} - LC_{95}$  ranged 0.05139 – 0.13976 and 0.02487 – 0.09126 after 24 and 48-h exposure respectively. These values indicated high susceptibility of these larvae against this formulation. In addition, no pupae emerged against 10, 1 and 0.1 ppm, while pupae formation reduced by 64, 57 and 46% against 0.01, 0.001 and 0.0001 ppm. Different concentrations were also evaluated in a semi-field assay ranged from 2.50 to 0.05 mg/L from July-August 2007. One hundred percent mortality was observed against 2.50 to 0.2 mg/L, 24 h post-exposure. However, *Bti* activity was higher in August as compared to July.

# 148 Response of phlebotomine sand flies to light-emitting diodes in southern Egypt

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Centers for Disease Control and Prevention light traps were modified for use with light emitting diodes and compared against a control trap (incandescent light) in an attempt to determine the effectiveness of blue, green, and red light against standard incandescent light routinely used for sand fly surveillance. Light traps were baited with dry ice and rotated through a  $4 \times 4$  Latin square design during May, June, and July 2006. Trapping occurred in the village of Bahrif, near Aswan on the east bank of the Nile River Valley in southern Egypt. Three repetitions netted 2,366 sand flies over 12 trap nights. *Phlebotomus papatasi* comprised 90% of total trap collections with three other species collected in small numbers. Over half (54.5%) of all sand flies were collected from red light baited traps and significantly more sand flies were collected from red light baited traps than from blue or green baited traps (p < 0.05). Red light baited traps collected more than twice as many sand flies as control traps and  $> 4 \times 10^{-5}$  more than blue and green light-baited traps. Red light is preferred when surveying where *P. papatasi* is the predominant sand fly species.

# 149 Preliminary studies of cultured and uncultured midgut of *Anopheles darlingi* from Amazon region using 16S rRNA gene sequencing

Thania Virgínia Guaycurus, guaycuru@centroin.com.br, L. K. Pereira, R. G. Lima, S. R. Cordeiro, E. N. Asuncao, W.P. Tadei, and S. Astolfi-Filho

In the Amazon region, malaria represents ~500,000 cases per year. An alternative strategy for vector control could be to observe the genetic variability in the vector populations or use paratrangenesis where bacteria are modified genetically to produce anti-parasitic factors and then introduced into the insect midgut. In this preliminary investigation, 2 different detection pathways were used to screen for bacteria in Anopheles darlingi mosquito midguts, 1 culture independent and 1 culture dependent. Indoor-resting, blood-fed female of An. darlingi were caught in Puraquequara, Manaus. Individual midguts were smashed in 50 µl of sterile water. The midgut suspension was streaked on Luria-Bertani agar (LB) plates and incubated for 48 h at room temperature. DNA templates were prepared by boiling or a bacteria colony or direct midgut suspension (10µl) for 10 min in a Tris-EDTA buffer (20mM Tris, 2mM EDTA, 1% Triton). The 16S rRNA primers used for the PCR experiments were designed in the Laboratory of Recombinant DNA - UFAM. The PCR products for the sequencing were prepared using ExoSAP-IT protocol. The sequencing was performed in capillary sequencer. For the preliminary identification, the 16S rRNA gene sequences were analyzed in BLASTn (http://ncbi.nih.gov/BLAST). In this pilot investigation our findings showed a great diversity of bacteria in cultured and uncultured midgut of An. darlingi as listed: Chromobacterium violaceum, Aquitalea magnusonii, Chromobacterium haemolyticum, Gulbenkiania mobilis, Vogesella indigofera, Laribacter hongkongensis, Paludimonas yongneupensis, Enterobacter spp., Enterobacter cloacae, Enterobacter endosymbiont, Enterobacter cowanii, Escherichia senegalensis, Salmonella spp., Salmonella typhi, Klebsiella pneumoniae, Aquitalea magnusonii, Vogesella sp., and Beta proteobacterium

#### 150 Genetic variability and phylogeography of Aedes aegypti populations from Brazil

Vera Margarete Scarpassa, vera@inpa.gov.br, Tatiana Bacry Cardoza, and Rubens P. Cardoso Jr.

Aedes (Stegomyia) aegypti is the most important vector of dengue and urban yellow fever viruses in the Americas. We conducted a population genetics study in 14 samples of Ae. aegypti from the north, southeast, northeast, and central regions of Brasil, including 7 states. An 852 bp region of the Cytochrome Oxidase I (COI) gene of mitochondrial DNA was used to estimate the population structure and dispersal rates, via gene flow, among the samples. A total of 163 individuals were sequenced. Ten haplotypes were observed. Genetic differentiation showed a large range (Fst = -0.085 to 0.992), with corresponding Nm values from Inf. to 0.004, which was not statistically significant for the most comparisons. From the 14 samples analyzed, Taubaté (State of São Paulo) showed the highest differentiation. Isolation by distance analysis indicated that the gene flow is related to geographical distance. Two haplotype groups were found, separated by 8 fixed mutations, suggesting the presence of 2 genetically distant lineages within Brasil. These data suggest that 2 or more introductions of Ae. aegypti may have occurred in Brasil, which may reflect in differences in vector competence and vector control measures for these populations. This research was supported by FAPEAM and MCT/INPA, Brasil.

#### 151 Influence of climatic change on mosquito development and mosquito-borne diseases in Europe Norbert Becker, kabs-gfs@t-online.de, Katarzyna Rydzanicz, and Bjoern Pluskota

In Europe, mosquitoes have a strong impact on life quality of humans, tourism and economic development and can

also threaten public health by transmitting mosquito-borne diseases. In this paper, the consequences of climatic change on the development of mosquitoes and their control as well on emerging diseases are discussed. The comparison of temperature development in 3 time periods as 1952-1966, 1967-1981, and 1982-2006 in Central Europe will serve as basis for the changing scenario related to population dynamics, abundance, and phenology of mosquitoes. Furthermore, the risk for mosquito-borne diseases such as West Nile fever will be discussed based on the adaptation capacity of Aedes albopictus to climatic change. Aedes albopictus has been spreading world-wide for decades due to its high adaptation capacity to various climatic conditions. Potential invasion places for Ae. albopictus have been investigated such as plant markets which offer lucky bamboo, stock piles of imported scrap tires, cargo harbour, railway stations, service areas along tourist routes, and cargo traffic from countries where Ae. albopictus occurs. The recent outbreak of chikungunya fever in Italy and the vector capacity of anophelines in Central Europe for the transmission of *Plasmodium falciparum* causing malaria tropica will be highlighted.

#### 152 From oil and Paris green to DDT: A brief history of the origins of chemical control Gordon Patterson, patterso@fit.edu

This paper traces the development of chemical means of control in the 19<sup>th</sup> and early 20<sup>th</sup> centuries up to the discovery of DDT in the early 1940s. In America, individuals noted oil's effectiveness as a larvicide in the 18<sup>th</sup> century. A century elapsed, however, before oil became widely used. In the late 1890s, L.O. Howard's publication of a series of articles on his experiments using oil against mosquitoes sparked new interest in chemical control. In 1913, the USDA established a research station at Mound, Louisiana, to explore chemical means of control. Barber and Hayne's (USPHS) discovery of the effect of Paris green against anopheline mosquitoes in the early 1920s led to a series of pioneering experiments by Willard King at the Mound laboratory. Simultaneously, a new generation of researchers such as Joseph Ginsburg (Rutgers) began to explore new means of chemical control and distribution techniques.

#### 153 Introduction to Insect Repellent as a Public Health Strategy Symposium

Emily Zielinski-Gutierrez, ebz0@cdc.gov

This symposium addresses the role that insect repellent can play in efforts to reduce vector-borne disease in the US. It is intended to both provide technical information useful to mosquito abatement districts in answering questions from citizens and the media, as well as to provoke discussion about the challenges to increasing consumers' use of this important public health strategy.

#### 154 How to understand the results of a mosquito repellent test

Donald Barnard, don.barnard@ars.usda.gov

This talk will discuss laboratory and field methodologies for the determination of repellent activity, including a brief discussion of sources of variation in such tests. The speaker will provide insight regarding how to understand and evaluate repellent test protocols and the data that are produced as a result.

#### **Communication of repellent efficacy** 155

Candace Brassard, Brassard.Candace@epamail.epa.gov

The EPA's Office of Pesticide Programs is undertaking a new initiative to improve public health by enhancing consumer understanding of the efficacy of skin-applied insect repellents and risks of vector-borne diseases. As a response to increasing news about vector-borne diseases, the public is becoming increasingly aware of repellents as an essential tool for both disease avoidance and nuisance control. But, there is confusion about the variable information found on insect repellent products. For example, while 50% of insect repellents have protection time for mosquitoes on the product label, only 20% of products have protection time for ticks. The consumer is receiving varied, and in some instances a complete lack of, information on the efficacy of these products. EPA's goal is to use

simple communication tools to enhance consumer understanding of the efficacy of these products by following the principles of accurate, clear, and consistent information transmittal. Initial products, including a website with information on specific products and efficacy for ticks and mosquitoes and efforts to develop clear and consistent ways to express efficacy on product labels, will be discussed.

# 156 What consumers should know about repellents: Considerations for product selection and recent improvements to product formulations

Keith Kennedy, keithkennedy@wi.rr.com

M. Keith Kennedy, Ph.D., of Science Strategies LLC, is an industry consultant and entomologist. Formerly director of S.C. Johnson and Son's Insect Control Product Development Division, Dr. Kennedy was associate professor of entomology (tenured) at Michigan State University. His presentation will provide an overview of items that the public should be aware of in regard to selecting and using repellents.

# 157 What role for repellents in public health? What else should we be doing?

Emily Zielinski-Gutierrez, ebz0@cdc.gov

This talk will address key factors in consumers' patterns of repellent use, including reasons cited for non-use. The role of repellent use as a public health strategy against vector-borne diseases will be discussed. The author will review the fact that we've seen relatively little shift in overall US rates of repellent use in recent years (at least according to available data) and prompt discussion on strategies for the future.

# 158 Effects of naled on nontarget insects during biting fly control operations at Parris Island, SC Mark Breidenbaugh, mark.breidenbaugh@youngstown.af.mil, and James Clark

The US Air Force uses a modified C-130H with flat-fan nozzles for ultra-low volume aerial spray applications. Nuisance and vector populations of adult saltmarsh mosquitoes and biting midges at the Parris Island Marine Corps Recruit Depot, SC, are managed using aerial spray applications. This study measured diversity and abundance of nontarget insects by monitoring populations before and after naled (Dibrom®) applications. Malaise and colored pan traps were used to passively sample populations of flying insects during 24-h intervals and compared with a no spray area. Application parameters, meteorology, as well as effects on biting fly populations and nontarget insects are discussed.

### 159 Efficacy evaluation of aerial adulticiding in Sacramento County, CA, 2007

Paula A. Macedo, pmacedo@fightthebite.net, Gary W. Goodman, and David A. Brown

The Sacramento and Yolo Mosquito and Vector Control District (SYMVCD) conducts surveillance and control of mosquitoes in Sacramento and Yolo Counties in CA. The District monitors adult mosquito abundance with American light traps, gravid female traps, and Mosquito Magnet Traps. We also use encephalitis virus surveillance (EVS) traps baited with CO<sub>2</sub> and gravid female traps to capture live mosquitoes to test for WNV and monitor minimum infection rates (MIR). With the increase in numbers and infection rates of *Culex tarsalis* and *Cx. pipiens* mosquitoes to 10.9 and 8.01 respectively, the District made the decision on July 26 to conduct aerial spraying of Evergreen. EC-60-6, over about 215 km2 in the northern area of Sacramento County on the nights of July 30, 31, and August 1. At the same time, we received notification of the first human case in the area. We conducted pre and post-trapping inside and outside the spray zone to evaluate mosquito abundance and infection rates after the spraying events. Results show a decrease in abundance of Cx. *tarsalis* and Cx. pipiens, a decrease in the percent positive mosquito pools collected, and a decrease in the minimum infection rate for Cx. *tarsalis*, which may have had an effect in decreasing West Nile virus transmission.

#### 160 Assessing ULV aerial application of Permanone 30-30: Efficacy and impact

He Zhong, zhongh@knology.net, Ed Hunter, Max Feken, and Cate Brock

Aerial ultra-low-volume (ULV) application of mosquito adulticides is widely used to control adult mosquitoes and reduce mosquito-transmitted diseases. Permethrin products are registered mosquito adulticides and are routinely used for ground ULV application. However, the aerial application of permethrin is not permitted in Florida because of concerns about toxicity toward aquatic non-targets. For example, the  $LC_{50}$  for rainbow trout and blue gill is 3  $\mu$ g/L and 5  $\mu$ g/L, respectively, as documented in laboratory 96 h fish bioassays. Also, there are concerns that large-scale aerial ULV application of permethrin may result in increasing aquatic non-target mortality. The FL Department of Agriculture and Consumer Services (FDACS) funded this research to examine the impact of permethrin on mosquitofish and the control efficacy against caged adult mosquitoes. The objectives of our research are to: 1) measure permethrin residue ground deposition, 2) determine permethrin airborne concentration, 3). measure permethrin deposition into the water in fish holding cage, 4) conduct fish bioassays to assess the water toxicity resulted from the residue deposition in field, 5) evaluate mosquito control efficacy.

# 161 Environmental concentrations of resmethrin in salt marshes: Measurements and implications David J. Tonjes, david.tonjes@stonybrook.edu, and Bruce Brownawell

In conjunction with a project to revise and optimize a mosquito control program for Suffolk County (Long Island), NY, 2 operational applications of resmethrin (Scourge<sup>®</sup>) were monitored in a mosquito control ditch in a salt marsh. Some of the aqueous samples were analyzed using high performance liquid chromatography coupled to

time-of-flight mass spectroscopy (LC-TOF-MS) by Stony Brook, and others were analyzed by the US Geological Survey using gas chromatography quadruple mass spectrometry (GC-MS). Sediment samples were analyzed by Stony Brook using GC-MS. The data show that resmethrin, while initially exceeding 100 parts per trillion (ppt) concentrations immediately after the applications in the water column, quickly decrease to less than ppt levels within hours. Although particle scavenging appears to be part of the mechanisms causing this rapid decline in the water column, resmethrin was not detectable in sediment samples. The data strongly suggest that non-target impacts to aquatic organisms from mosquito control applications of resmethrin are unlikely to occur, unless the initial concentrations are immediately fatal.

#### 162 A ground ULV field trial using Duet® against Culex salinarius in Louisiana

Scott Willis, swillis@cppj.net, and Scott Harrington

This paper will focus on a ground ULV field trial that was conducted to determine the efficacy of Duet<sup>®</sup> at a rate of 0.0012 lbs (sumithrin) and 0.0003 lbs (prallethrin) active ingredient per acre. In May 2007, this trial was conducted in Lake Charles, LA against *Culex salinarius*. A standard, replicated ground ULV open field caged protocol was used to conduct this trial. The results showed that 99% knockdown was recorded at 1 h and at 24 h for mortality.

### 163 The performance of AquaHalt® in the aerial operational program of Grant County Mosquito Control District #1

Jim Thompson, gcmcd@nwi.net

In 2007, AquaHalt<sup>®</sup> was used in the aerial adulticide program in the Grant County, WA, to control adult mosquito populations. This paper will discuss the droplet dynamics during the characterization of the aircraft. In addition, an open field caged trial was conducted at the operational application rate to determine the efficacy of AquaHalt<sup>®</sup> against *Coquillettidia perturbans*. Results of this trial and other aspects will be discussed.

#### 164 A review of 2007 field trial results for Duet®

Bill Jany, bill@clarkemosquito.com

During 2007, a number of  $DUET^{\oplus}$  operational and open caged field trials were conducted across the country. This presentation will discuss  $DUET^{\oplus}$  applications made with ground and aerial equipment and the results obtained against several mosquito species. Specific information on calibration, characterization, and efficacy validation will be reviewed.

### Preliminary results of Duet<sup>®</sup> adulticide against *Aedes albopictus* (Skuse) in Middlesex County, New Jersey Glenn A. Levinson, glevinson@middlesexmosquito.com, and Deepak Matadha

Within the past 2 yr, there has been a rapid increase in the abundance of *Aedes albopictus* (Skuse) (Culicidae: Diptera) in several areas of NJ. In 2007, the Middlesex County Mosquito Extermination Commission conducted pre and post-application landing counts to evaluate the efficacy of Duet<sup>®</sup>, a dual-action adulticide for controlling *Ae. albopictus*. Duet<sup>®</sup> applied using ground ULV equipment at the rate of 0.75 oz/acre provided good control of *Ae. albopictus*, producing significant reduction of adults after 24 h post-application. The preliminary results of the field test and the potential integration of Duet<sup>®</sup> into the Commission's adult mosquito control program will be discussed.

### Field trials in the northeast: AquaAnvil®, Duet®, and a new learning experience Robyn Januszewski, frankrenick@clarkemosquito.com, and Emily Sullivan

As concern grows over the environmental effects of and insects' resistance to pesticides, it is in the best interest of mosquito control districts to investigate new products that can be added to supplement current control methods. In 2007, the Northeast Massachusetts Mosquito Control and Wetlands Management District (the District) was invited to participate in the testing of two new products, AquaAnvil® and Duet®, by Clarke Mosquito Control. District personnel were responsible for nearly all aspects of the trials from site selection, and collecting and caring for the mosquitoes to monitoring the weather and running the trials. Not only did the trials provide the District with a unique learning experience, they also provided Clarke Mosquito Control with efficacy data on the 2 products.

#### 167 Introduction to the Student Paper Competition

C. Roxanne Connelly, crr@ufl.edu

The AMCA Student Paper Competition includes undergraduate and graduate students conducting research in medical entomology, public health, and related fields. In 1988, AMCA held the first student paper symposium at the annual meeting to focus on the contributions of AMCA student members. The Board of Directors approved the first student paper competition for the 1989 meeting in Boston, MA. The Student Competition remains an important part of the AMCA's annual meeting program.

### Mosquito management and risk: Environmental concentrations of insecticides and their meaning Jerome J. Schleier III, jerome.schleier@myportal.montana.edu

We conducted environmental fate studies on adulticides applied using aerial and ground ultra-low-volume (ULV) equipment. Our results suggest that the amounts of adulticide deposited on the ground and in water are lower than what has been estimated by previous exposure and risk assessments.

### 169 Relationship between kdr mutation and resistance to pyrethroid and DDT insecticides in *Anopheles gambiae* Lisa Reimer, ljreimer@ucdavis.edu

Two mutations in the voltage-gated sodium channel gene confer knock down resistance (kdr) to pyrethroids and DDT in *Anopheles gambiae*. We analyzed the frequency and relationship between kdr genotypes and resistance to Type I and II pyrethroids and DDT in both the Forest-M and S molecular forms of *An. gambiae* in Cameroon. Implications for malaria control and the reintroduction of DDT are discussed.

#### 170 Update report of West Nile virus surveillance in Lubbock County, TX

Virna L. Saenz, virna.saenz@tiehh.ttu.edu

West Nile Virus (WNV) surveillance efforts have been continued since its first appearance in Lubbock County, TX in 2002. Here we report an update of WNV mosquito surveillance results in Lubbock County during the period 2004-2007. Adult mosquitoes collected represent seven genera; *Culex tarsalis* and *Aedes vexans* were the most common species collected. Mosquitoes were pooled and screened for WNV using the VecTest<sup>TM</sup> and reverse transcriptase polymerase chain reaction.

### 171 Analysis of a local mosquito population: A spatial comparative analysis of larval and adult mosquitoes Marshall D. Lipps, mlipps@comosquitocontrol.com

Current policy of the Boulder County Mosquito Control District seeks to reduce adulticide applications focusing rather on targeted larval applications. This study will look at spatial associations of larval and adult populations to determine if key sources of mosquitoes can be identified to explain historically high adult populations in order to aid targeting and increase efficacy of larval control activities to ultimately reduce disease transmission, human annoyance, and chemical adulticide applications.

### 172 Modeling Culex tarsalis abundance using meteorologic and edaphic predictors in California Christopher M. Barker, cmbarker@ucdavis.edu

The abundance and phenology of *Culex tarsalis* vary across the diverse ecological regions of California. Using Bayesian regression models fitted to a 10-year statewide surveillance dataset, we quantified the region-level responses of *Cx. tarsalis* to meteorologic and edaphic predictors. We also accounted for spatial and temporal dependence among trap counts, two aspects typically ignored in previous studies. Relative adult abundance depended on land use, human population density, and the intensity and timing of meteorological events.

#### 173 Weather influence on mosquito populations in West Texas

Walter A. Albeldano, walter.albeldano@tiehh.ttu.edu

Field studies were conducted in the city of Lubbock, Lubbock Co., TX during the years 2004 through 2007 to determine the seasonal pattern of mosquito population abundance and the potential effects of rainfall and temperature on the distribution of West Nile virus vectors. Mosquitoes were collected from early spring until late fall. Relationships between rainfall and temperature for various mosquito species, including *Aedes vexans* and *Culex tarsalis*, were observed.

### 174 Species composition of overwintering mosquito populations and biology of diapausing Culex pipiens pipiens Ary Farajollahi, farajoll@rci.rutgers.edu

We examined the seasonal phenology of overwintering populations of Cx. pipiens pipiens. Most diapausing mosquitoes collected were composed of Cx. p. pipiens (81.6%). Diapausing populations were primarily composed of inseminated (98.6%) and nulliparous (100%) females, with primary ovarian follicles in resting stage 1a, linear size of 50.8  $\mu$ m (95% CI = 40.3-61.3  $\mu$ m), and primary/secondary follicular ratio of <1.5 (95% CI = 1.2-1.7). One pool of overwintering Cx. p. pipiens was positive for West Nile viral RNA.

#### 175 Withdrawn

### 176 Development and efficacy of a novel fabric fiber impregnated with discriminatory insect repellent Emily Bick, emilybick@msn.com

Every thirty seconds a child dies of malaria. Long duration insect repellant cloth suitable for repelling disease vectors may be prepared by impregnating cotton fibers with polymethyl methacrylate microspheres which incorporate a discriminatory insect repellent. With a 95% confidence level, the experimental impregnated thread was shown to be more effective than similar thread treated with N, N-diethyl-m-toluamide ("DEET").

#### 177 Introduction to Mosquito Research Groups - What Are They up to Today?

Jeff Stivers, jeff@cmcd.org

The directors of 5 major resarch organizations will be presenting information on their personnel, current research interests, and future programs.

#### 178 Medical entomology at the University of FL's FL Medical Entomology Laboratory

Walter J. Tabachnick, wit@ufl.edu

#### 179 An update on research at the USDA/ARS Mosquito and Fly Research Unit

Gary Clark, gary.clark@ars.usda.gov

### 180 Research and teaching programs of the Division of Infectious Diseases of the William C. Gorgas Center for Geographic Medicine

Robert J. Novak, rjnovak@uab.edu

#### 181 Research initiatives of the University of California Mosquito Research Program

Gregory Lanzaro, gclanzaro@ucdavis.edu

### 182 Mosquito control research at Florida A&M University, Public Health Entomology Research & Education Center (PHEREC) in Panama City, Fl

John Smith, john.smith@famu.edu

#### 183 What happened to mosquito systematics?

Lawrence Hribar, lhribar@keysmosquito.org

The field of mosquito systematics appears to be dying slowly, and without much notice from the mosquito control community. Retirement, reallocation of efforts, reprioritization, and redirection of lives all may be contributing factors. The demise of the journal *Mosquito Systematics* has resulted in important papers being scattered into a number of other journals.

### 184 Molecular and cytogenetic evidence of three sibling species of the *Anopheles barbirostris* Form A (Diptera: Culicidae) in Thailand

Wej Choochote, wchoocho@mail.med.cmu.ac.th, Visut Baimai, Benjawan Tuetan, and Anuluck Junkum

Nine isoline colonies of *Anopheles barbirostris* Form A, derived from individual isofemale lines from Chiang Mai, Phetchaburi and Kanchanaburi, were established in our insectary at Chiang Mai University. All isolines shared the same mitotic karyotype (X1, X2, Y1). Molecular analysis of DNA sequences and PCR-products of ITS2, COI and COII regions revealed 3 distinct groups: A1 (Chiang Mai), A2 (Phetchaburi) and A3 (Kanchanaburi). Crossing experiments among the 3 groups exhibited strong reproductive isolation, producing low and/or non-hatched eggs, and inviable prallethrin and/or abnormal development of the reproductive system of F1-progenies. Asynaptic regions along the 5 polytene chromosome arms of F1-hybrid larvae clearly supported the existence of 3 sibling species within *An. barbirostris* Form A, provisionally named: Species A1, A2, and A3.

### 185 Cytogenetic and molecular evidence for two species in the *Anopheles barbirostris* complex (Diptera: Culicidae) in Thailand

Atiporn Saeung, wchoocho@mail.med.cmu.ac.th, Wej Choochote, wchoocho@mail.med.cmu.ac.th, Visut Baimai, Yasushi Otsuka, and Benjawan Tuetan

Seventeen isolines of *Anopheles barbirostris* derived from animal-biting females showed 3 karyotypic forms: Form A (X2, Y1) 5 isolines (Phetchaburi); Form B (X1, X3, Y2) 3 (Chiang Mai) and 8 (Ubon Ratchathani) isolines; Form C (X2, Y3) 1 isoline (Phetchaburi). All 17 isolines exhibited an average branch summation of seta 2-VI pupal skins 12.1-13.0 branches, which was in the limit of *An. barbirostris*. Of the 12 human-biting isolines from Chiang Mai province, 5 isolines showed Form B (X2, Y2) and 7 isolines exhibited a new karyotypic form designated as Form E (X2, Y5). All of 12 isolines had an average branch summation of seta 2-VI pupal skins 22.4-24.5 branches, which was in the limit of *An. campestris*. Thus, they were tentatively designated as *An. campestris*-like Forms B and E. Crossing between *An. campestris*-like Forms B and E yielded viable progenies, suggesting conspecific relationships. Reproductive isolation among crosses between *An. campestris*-like Form B and *An. barbirostris* Forms A, B and C strongly suggested the existence of these 2 species. The very low intraspecific variation of ITS2, COI and COII of *An. campestris*-like Forms B and E supported their conspecific relationship. The large sequence divergence of ITS2, COI and COII of *An. campestris*-like Forms B and E, and the *An. barbirostris* Forms A, B and C clearly supported cytogenetic and morphological evidence.

#### 186 Neoteny in the evolution of mosquito larvae

Thomas Zavortink, tjzavortink@ucdavis.edu

The history of the concept of neoteny and the development of its modern evolutionary definition are briefly reviewed. Neoteny is presently defined as a slower rate of developmental events (at any level: cell, organ, individual) in the descendant. The 4<sup>th</sup> instar larvae of some mosquitoes exhibit characteristics found in the 3<sup>rd</sup> or younger instars of related species. Neoteny has been postulated as the evolutionary explanation for this phenomenon. Early reports of neoteny in mosquito larvae are summarized and additional examples of

morphological characteristics of mature mosquito larvae that may owe their evolution to neoteny are presented. In addition to producing differences among related species, neoteny can also create analogous similarity in distantly related species. For those who believe that evolutionary relationships can be inferred only through the use of homologous characters, neoteny presents yet another challenge in the age-old problem of separating homologous from analogous similarity.

### 187 Identification of *Culex (Melanoconion)* species of the US using female cibarial armature and male genitalia Martin Williams, zoq9@cdc.gov

Species within the subgenus *Culex* (*Melanoconion*) are the primary enzootic vectors of Venezuelan encephalitis virus and Everglades virus. Adult females of this subgenus are often difficult or impossible to identify to species based on external morphological characters. The use of female cibarial armature and male genitalia allows for the identification of field collected adult specimens of *Culex* (*Melanoconion*). We describe and present illustrations of the cibarial armature and male genitalia for all US species and present keys to all US species using these characters.

#### 188 Genetics and morphology of *Anopheles maculatus* chromosomal form K

Damrongpan Thongwat, dthongwat@yahoo.com, Pradya Somboon, and Catherine Walton

The Anopheles maculatus group consists of 8 recognized members, i.e., An. maculatus, An. sawadwongporni, An. willmori, An. pseudowillmori, An. dravidicus, An. notanandai, An. greeni and An. dispar. In the taxon of An. maculatus, 3 chromosomal forms (B, E, K) have been described. Previous studies suggested that forms B and E are cytotypes of the species and they are indistinguishable in morphology. Few genetic characteristics of form K are known. The present study demonstrates, using crossing experiments and ovarian nurse cell polytene chromosomes of the F1 hybrids, that form K is genetically distinct from the other 2 forms. In addition, post-zygotic genetic incompatibility between form K and An. sawadwongporni, An. dravidicus, An. pseudowillmori and An. willmori is demonstrated. In all crosses, all hybrid males were sterile with atrophied testes and accessory glands, or partially sterile with abnormal spermatozoa. The hybrid females showed varying degrees of atrophied ovaries. The ovarian nurse cell polytene chromosomes of the F1 hybrid females displayed ~70% to almost complete asynapsis. Study on the adult and larval morphology form K reveals overlapping characteristics with An. sawadwongporni and An. notanandai. The present study supports previous suggestions that form K represents another species of the Maculatus Group.

### 189 Introduction and Overview to the Deployed Warfighter Protection Research Program: Finding New Ways to Vanquish Ancient Foes Symposium

Stanton E. Cope, Stanton.cope@ods.mil

The Deployed Warfighter Protection Research Program is a \$5 million/yr effort by the Department of Defense to find new pesticides, new application equipment, and new methods of personal protection for military personnel in operational settings. The Armed Forces Pest Management Board administers it. This symposium will summarize accomplishments and ongoing research efforts at several USDA laboratories, universities, industry, and military laboratories.

### 190 DWFP round up: Retrospect and prospects of the Research Program for Deployed War-fighter Protection from disease vectors

Graham White, gbwhite@ufl.edu, Stanton Cope, Daniel Strickman, and Richard Johnson

DWFP is an initiative to develop and validate novel methods to protect US military deployed abroad from threats posed by disease-carrying insects. This R&D program began in 2004, administered by the Armed Forces Pest Management Board, with recurrent \$5M/yr budget from DoD/DUSD(I&E). Sixty percent of funds have been committed to strengthening research with the medical entomology component of the USDA Agricultural Research Service. Facilities, accomplishments, and products of this ongoing activity will be summarized. Remaining funds are committed to competitive research awards to academia, industry, and military entomologists. An overview will be given of achievements from DWFP grants to ~40 scientists during 2004-2008, with examples of products that already benefit military deployments. This DWFP program sustains USDA research collaboration with DoD since WW2, improving capabilities for vector control, pesticide application, and personal protection, with spin-offs to veterinary and public health.

#### 191 Comparative biological risk research to protect deployed forces

Robert Peterson, bpeterson@montana.edu

The Comparative Biological Risk Assessment program at Montana State University has been involved in the DWFP program since 2005. We have been focusing on human-health and ecological risk assessments for vector-borne diseases and associated vector management strategies. Examples of risk assessments for vector management strategies include repellents, barrier sprays, insecticide-impregnated uniforms, and ground-applied adulticide space sprays. More recent research includes characterizing actual environmental concentrations and fate of adulticides.

### 192 University of FL evaluation of new technologies for protection of military personnel from flies Philip Koehler, Roberto Pereira, and Alexandra Chaskopoulou, andahask@ufl.edu

DWFP research at the UFL Urban Entomology Laboratory targeted the development of new insecticides for fly control. During the 3-yr research project, research on control of mosquitoes and flies developed from the initial screening of insecticidal active ingredients to field testing of new formulation and new application devices. We obtained several insecticides including pyrethroids, neonicotinoids, phenylpyrazoles, oxadiazines, and organophosphates and screened them against flies. We tested fly traps and light traps to optimize military usage of these non-chemical controls. A sprayable spot fly bait was evaluated, proved to be very useful for use by deployed troops, received a NSN (01-555-9369) and is available for use by military entomologists. New volatile compounds were tested against both flies and mosquitoes and were shown to be useful for control of these insects in confined areas. Insecticide-impregnated wool cords were shown to be the best material for delivery of the insecticides to flies, because there was an efficient acquisition of pesticide by the insects from wool cords, possibly because of the presence of natural oils. The grant supported 6 graduate students, 3 of them military entomologists, and 1 now working for the DWFP project.

### 193 Efficacy of volatile low molecular weight insecticides against *Aedes* and *Culex* mosquitoes Alexandra Chaskopoulou, andahask@ufl.edu, Sam Nguyen, Philip Koehler, and Michael Scharf

Toxicity bioassays were conducted to test a series of 30 volatile insecticides against 2 important disease-vectoring mosquitoes, *Aedes aegypti* (L.) and *Culex quinquefasciatus* Say. These insecticides were also tested previously using *Drosophila melanogaster* Meig. as a model dipteran species. Our findings across the 3 species provided comprehensive structure-activity profiles for the 30 insecticides. Interestingly, each species showed a unique structure-activity profile. Although the exact causes of these differences are not known, size and/or physiological differences between species may be at least partially responsible. These findings provided novel and important information regarding volatile insecticide toxicity that will refine future research questions and assist in the development of formal vector management programs.

### Juvenile hormone analogs and chitin synthesis inhibitors as rodent feed-throughs against sand flies (Diptera: Psychodidae)

Thomas Mascari, tmascari@agcenter.lsu.edu, Lane Foil, Ed Rowton, and Mark Mitchell

The efficacy of insect development inhibitors/insect growth regulators was evaluated as potential rodent feed-throughs for the control of immature stages of sand flies (*Phlebotomus papatasi* Scopoli). The development and survival of 2<sup>nd</sup> instar larvae fed feces from Syrian hamsters, *Mesocricetus auratus*, that had been fed a diet containing a juvenile hormone analogue (methoprene or pyriproxyfen) or a chitin synthesis inhibitor (diflubenzuron or novaluron) were evaluated. The feces of treated hamsters greatly reduced or prevented larval to adult development. The specific effects of each compound on the growth and development of sand fly larvae will be discussed. The results of this study suggest that a control strategy using rodent baits containing a juvenile hormone analog or chitin synthesis inhibitor to control phlebotomine sand flies that live in rodent burrows and feed on rodent feces may be possible. Since the rodent reservoirs and vectors of *Leishmania major* live in close association in many parts of the Middle East, the control of transmission of the agent of zoonotic cutaneous leishmaniasis also may be possible.

### 195 The biological activity of a novel class of insecticides, sterol carrier protein inhibitors (SCPIs) Ryan Larson, rtlarson@wisc.edu, and Que Lan

Insects are unable to synthesize cholesterol de novo and must rely on dietary sources of cholesterol. The transport of lipids is aided by the sterol carrier protein-2 (SCP-2). Several sterol carrier protein inhibitors (SCPIs) have been discovered through high throughput chemical screening. SCPIs represent a class of novel insecticides. Biological activities of SCPIs were evaluated in several species of mosquitoes, the  $LC_{50}$  ranged from 0.9ppm to 11ppm depending on the species and stages when SCPIs were applied.

#### 196 Etofenprox: New active ingredient for control of adult mosquitoes

David Malone, dmalone@e-adapco.com, Todd Gwinn, and Allen Wooldridge

Etofenprox is a non-ester pyrethroid identified in the early 1980s by Mitsui-Toatsu and developed initially for the control of crop insects in Japan and Europe. First registrations were achieved in the early 2000's, primarily as consumer products such as dog & cat spot-ons, aerosols, foggers. Now licensed by Central Life Sciences and distributed by ADAPCO for mosquito control in the USA, etofenprox ULV was first applied in Georgetown, SC, June 2005, against *Ochlerotatus taeniorhynchus* and *Oc. sollicitans*. Subsequent trials in the US and Caribbean islands have achieved good rates of knock-down and effective kill of a wide range of mosquito species. EPA registration of the US label is anticipated soon for the ULV adulticide etofenprox.

#### 197 The virtual laboratory at USDA ARS

Daniel Strickman, daniel.strickman@ars.usda.gov

The Veterinary, Medical, and Urban Entomology national program of the USDA ARS has broad expertise in entomology ranging from genetics to trapping technology. The Deployed Warfighter Protection program has stimulated the program to integrate its efforts across laboratories in order to produce products useful for deployed

military personnel. The basic steps involved are discovery, evaluation, development, and industrial association. Five laboratories and some 30 people work together to apply basic science to applied problems, culminating in commercially produced products. The administrative infrastructure within ARS has been invaluable in supporting these efforts, including the ability to contract work, to patent inventions, and to establish legal relationships with industry.

#### 198 Sentinel GIS: Simple, yet powerful, GIS tools for mosquito control operations

Ryan Pierson, ryan@elecdata.com, and Mike Swan

Using industry-leading technology, mosquito control professionals are implementing simple, yet powerful, GIS applications for mosquito control. Built upon the core ESRI GIS technology, the industry leader in GIS, the B&G Sentinel GIS tools are developed and being customized to address mosquito adulticide, larvicide, and surveillance requirements. These solutions provide the mosquito control professionals with the ability to easily update maps, track chemical usage, intelligently design inspection and treatment schedules, and generate all necessary reports. This solution is completely customizable to meet the requirements of each mosquito control agency and easily integrates with existing database applications already in use. This presentation will describe these easy-to-use applications for mosquito control operations and highlight the industry leading technologies used to create the B&G Sentinel GIS products. Included will be real-world example data and reports from current users of this technology. Highlighted will be implementation details, including costs, and also realized time/cost savings by using these tools from current users. These powerful solutions are developed around the core ESRI GIS technology for the office and the field, and also integrate industry-leading technology from Trimble Navigation, Juniper Systems, and Electronic Data Solutions.

#### 199 Web Map connects citizens, staff, and data

Nancy Read, nancread@mmcd.org, Brian Fischer, Mike McLean, and Jon Peterson

Citizens call the Minneapolis/St. Paul-area Metropolitan Mosquito Control District every year to ask if particular wetlands in their area are being treated to prevent mosquito problems. In 2007, we launched a web-based wetland look-up application that provides a simple interface to allow users to check 2 yr of inspection and treatment history for any of the 70,000 wetlands in the District. The web application uses public aerial photos and other web map service layers in an interface built with a free open source software package called GeoMoose and using the open source MapServer engine. Inspection and treatment data are updated nightly. The intent was to make map and treatment data easily available to all staff at our 7 facilities and to the public at large. Citizens began finding the site early in the year and the site was featured in TV coverage on the District. To get information on GeoMoose go to http://www.mmcd.org/treatentrypage.htm to view the site, or http://www.geomoose.org/moose.

### 200 New software release adds significant enhancements to the Vector Control Management System (VCMS) Rob Olson, robolson@clarkemosquito.com

Advanced GPS/GIS mapping capabilities and the use of cutting-edge technologies are just a few of the enhancements of the new Vector Control Management System software release that can help optimize your operational program and decision-making.

#### 201 Spray vehicle real-time meteorological data observation and collection

Jacob Mundt, info@larvasonic.com, and Herb Nyberg

Meteorological conditions impact chemical spray applications in many dimensions. Both real-time meteorological data as well as forecasts can enhance mosquito control programs. For instance, identification of temperature inversions can be beneficial to some type of applications. Knowing wind direction can enhance the drift patterns. Many mosquito control districts have made attempts to monitor meteorological conditions during spray operations but they required the vehicle to stop and manually record data. The first integrated GPS and ultrasonic anemometer/wind vane was developed for marine applications. It is attractive for mosquito control because the acoustic sensors have no moving parts and greatly improve system reliability and the integrated GPS allows for conversion of apparent wind into true wind speed and direction. Some chemical labels require that wind speeds be monitored at the point of application. Ada County ID Mosquito and Weed Control evaluated this technology and results are presented.

### 202 MosquitoMap: A new web mapping site for mosquito species distribution and vector-borne disease risk assessment

Desmond H. Foley, foleydes@si.edu, and Richard C. Wilkerson

We introduce MosquitoMap, an online geospatially referenced clearinghouse for mosquito species collection records and species distribution models. Users can pan and zoom to anywhere in the world to view the locations of past mosquito collections and the results of models that predict the geographic extent of individual species. Collection records are searchable and downloadable, users can map and upload their own georeferenced collection data or distribution models, and all contributions are fully attributed. MosquitoMap includes a vector-borne disease risk analysis tool called the Mal-area calculator. This tool combines models of mosquito vector distribution and pathogen suitability to determine the extent of the area where these organisms co-occur. The vectorial importance of each species is adjustable, the area of interest is resizable, and different locations can be compared to determine

relative risk. MosquitoMap is designed to preserve and make available the results of past collecting and distribution modeling activity. MosquitoMap has a starter database of over 65,000 collection records for 1,176 taxa, and about a dozen species distribution models. However, the utility of MosquitoMap will increase as more records are added. Contributions are encouraged, especially from individuals and organizations with digitized, georeferenced records and those involved in ongoing mosquito surveillance.

### 203 Using satellite climate measurements to understand dynamics of mosquito populations in the US Seth C. Britch, seth.britch@ars.usda.gov, Kenneth J. Linthicum, and Assaf Anyamba

Precise, accurate predictions of mosquito densities and distributions would be significant assets to reducing the severity of existing or emerging mosquito-borne disease events in the US. We are examining changes in populations of mosquito species from long-term surveillance records provided by mosquito and vector control districts and public health agencies across the country. Mosquito population dynamics are compared to satellite climate measurements to find environmental signals that tend to precede anomalously high or low populations. We present examples of the promising outcomes of these comparisons, and describe how they could be used as key features of national mosquito-borne disease surveillance and control programs. We highlight the 2006/2007 outbreak of Rift Valley fever in the Horn of Africa and explore how predictive mosquito population data could have been used to identify vulnerable nodes of contact between the US and eastern Africa and efficiently target protective measures.

#### 204 The Beech King-Air: A multi-use aerial spray platform for the 21st century

Pamela C. Jacobson, JacobsonP@hillsboroughcounty.org, Brian Lojko, and Wayne Daniels

In partnership with Embry-Riddle Aeronautical University's Eagle Works engineering team and Pasco County Mosquito Control District's pilot/design engineer, Hillsborough County Mosquito and Aquatic Weed Control has developed a state-of-the-art high-pressure aerial adulticide system with an external pesticide tank for our King Air C90 twin turbine airplane. With the exception of a small amount of wiring and hardware, the entire system can be easily removed within 4 h, returning the airplane to normal category as a passenger transport aircraft. Even in FL, where mosquito control is a nearly year-round activity, aerial application aircraft are often subject to long periods of disuse, during which they can develop corrosion and decay unless regularly exercised. Keeping this aircraft "dual category" provides opportunities for regular public works support activities during slow periods, while maintaining a valuable asset capable of responding aggressively and decisively to a mosquito-borne disease outbreak.

#### 205 Development of a spray system for an autonomous helicopter

W. Clint Hoffmann, choffmann@tamu.edu, and Yanbo Huang

A spray system has been developed for placement on a fully autonomous helicopter. After programming the helicopter to fly a spray route, the spray system can be programmed to come on and off only when the helicopter is within a predefined spray block. The work presented will detail the operational parameters of the helicopter and spray system, as well as, the possible uses of these systems in vector control applications.

### The evolution of aerial application in Butte County MVCD, CA: Upgrading our aerial assets with advanced flight guidance systems

Jim Camy, bcmvcd@nvwisp.com, and Del Boyd

Butte County MVCD headquartered in Oroville, CA uses 3 fixed-wing aircraft to apply larvicides and, when necessary, adulticides to prevent or reduce the risk of West Nile virus and other mosquito vectored diseases. During 2007, the district purchased an advanced flight guidance system and on-board meteorological instrumentation to upgrade our aerial adulticide operation and improve the efficacy of our applications. This presentation reviews the build-out, training, and integration of this equipment into our program. The lessons learned from this experience may help other districts considering similar upgrades.

### 207a Introduction to the Aerial Treatment Guidance/Tracking GPS – Experience from the Field Symposium Nancy Read, nancread@mmcd.org

Aerial treatment guidance and tracking systems can be a great tool for mosquito control operations and also can present interesting challenges. Following a brief introduction to the basic concepts, a panel of aerial system users will describe their systems, how they use field inspection data and prepare information on areas to treat, what the pilots see in-flight, how tracking information is handled, and general comments on what has worked well and what hasn't. Discussion will include costs and how users balance their investment of time and resources with benefits such as treatment accuracy and defensible records.

#### 207b Larviciding in Minneapolis/St. Paul, MN

Nancy Read, nancread@mmcd.org

The Metropolitan Mosquito Control District has used AgNav Guia systems in helicopters for larval control for the past year. Field inspection results by site number are compiled at the end of the day and linked with site boundaries in MapInfo GIS to produce a shapefile of sites to treat, which is uploaded to AgNav units the next morning. Pilots use boundaries to verify site location and may use swath guidance on larger sites. Tracks are downloaded,

converted, and reviewed in MapInfo. This system has helped staff and pilots keep track of treatments, which may include 100 sites per pilot per d.

#### 208 Aerial larviciding of spring Aedes mosquitoes in Saginaw County, MI

Randall Knepper, randy@scmac.org, Albert Schiffer, and Michael Schiffer

Saginaw County Mosquito Abatement Commission has used Hemisphere's (Satloc) MapStar<sup>TM</sup> aerial guidance and tracking systems for 13 years for larviciding spring *Aedes* mosquitoes with fixed wing aircraft. Field inspections are compared with human population distribution to select areas for spring treatment. GIS shape files for woodlands to treat, woodlands that will not be treated, and no-spray exclusions zones are loaded into the MapStar software in aircraft and ground crew computers. Real-time tracking allows ground staff to monitor and evaluate aircraft during larviciding. Treatment tracks are downloaded and converted for use in ArcView GIS with county data to produce permanent records of larviciding application. This system has helped focus resource use, improve treatment coverage, and answer questions from the public.

#### 209 Larviciding in Winnipeg, Manitoba

Taz Stuart, tstuart@winnipeg.ca

Winnipeg's aerial program has used Trimble guidance and tracking systems for 8 yr in 4 helicopters for larvicide work. Field site pre-checks are called in to a helicopter dispatcher and a list of sites to treat is prepared for the helicopter for the current or next day. GIS files for treatment areas are uploaded using "Bugbase" and "Proximity Pilot" software, and no-spray areas and buffer zones are shown on screen. Treatment tracks are downloaded and displayed in ARCGIS and in MapInfo with associated municipal data. A similar system is used for ground-based adulticiding. This system has helped maintain quick response to treatment needs while respecting no-treatment requests.

#### 210 Larviciding and adulticiding in the Keys, FL

Ralph DePalma, rdepalma@keysmosquito.org, and Amy Sargent

Keys Mosquito Control has used Adapco Wingman guidance and tracking systems for 2 yr for aerial adulticide and larvicide work in both fixed wing aircraft and helicopters. GIS shapefile areas are uploaded and no-spray areas designated that automatically turn sprayers off. Helicopter larviciding operations use a passenger as guide in addition to the Wingman system. Treatment tracks are downloaded to ArcGIS. This system has helped improve treatment results and reduce insecticide use, especially in sensitive areas.

#### 211 Larviciding and adulticiding in Manatee County, FL

Mark Latham, manateemcd@aol.com

Manatee County has used aerial guidance systems since 1994 and currently uses Wingman systems in both helicopters for larvicides and fixed wing aircraft for adulticiding. Field check results are used to map treatment areas in MapInfo GIS, converted in Adapco's Skytracker software, and uploaded to the Wingman units. Records of tracks with spray on/off data, flow, and weather are downloaded to Skytracker and converted back for use in MapInfo. This system has improved treatment coverage accuracy, especially with addition of weather variables and drift offset modeling for adulticides, as well as providing defensible records. Tower maps aid safety for pilots.

#### 212 Larviciding and adulticiding in Chatham County, GA

E. Scott Yackel, esyackel@chathamcounty.org

Chatham County has used flight guidance for many years, including AgNav 2 units since 1999 and AgNav Guias since 2006, in both fixed wing aircraft and helicopters. For larviciding, areas to be treated are designated by number and field staff will call helicopters with additional target numbers during the 3-d flight period after a rainfall. For adulticiding, areas are described by landmark boundaries and pilots decide the best flight path to cover target area. Pilots see progress on moving map screens and tracks are downloaded to ArcGIS and overlaid on county maps and digital photos. Swath guidance has become an important tool for pilots and recorded tracks have helped diffuse citizen concerns.

#### 213 Aerial larviciding and adulticiding in Butte County, CA

Jim Camy, bcmvcd@nvwisp.com

Butte County has used SatLoc for many years in fixed wing aircraft for larviciding and adulticiding and, during the past year, used the AgNav/FlightMaster system for adulticiding. Field staff mark larval locations in Google Earth and e-mail map and coordinates to office where information is given to the pilot. Spray tracks are stored for later review in ArcView. Adulticide areas are set up in FlightMaster, including boundaries of organic exclusion areas. Spray tracks can be reviewed in FlightMaster or overlayed on a GIS or Google Earth. The FlightMaster system has allowed much more accurate treatments around certified organic agricultural fields by using on-board meteorological data gathering equipment to insure ULV applications hit the target area and avoid nontarget fields.

#### 214 Insights into Anopheles (Nyssorhynchus) (Diptera: Culicidae) species from Brasil

Maria A. M. Sallum, masallum@usp.br, Sandra Nagaki, Gabriel Laporta, and Cecilia L. S. Santos,

Anopheles (Nyssorhynchus) benarrochi s.l., An. (Nys.) oswaldoi s.l. and An. (Nys.) konderi s.l. collected in Acrelândia, State of Acre, Brasil, were identified based on morphological characters of the male genitalia, 4<sup>th</sup> - instar larvae, and pupae. Morphological variations were observed in the male genitalia of these species when compared to specimens from Acrelândia and other localities in Brasil. DNA sequence data from the second nuclear internal transcribed spacer region (ITS2) of individuals identified, based on male genitalia characteristics, showed that the different morphological forms of An. benarrochi s.l. are cospecific, however, distinct from An. benarrochi B from Colombia. Anopheles konderi s.l. and An. oswaldoi s.l. may comprise two undescribed species both largely misidentified as An. (Nys.) oswaldoi s.s. (Peryassú) throughout Brasil. Diagnostic morphological characteristics of the male genitalia are provided to distinguish An. benarrochi s.l., An. oswaldoi s.l. and An. konderi s.l. from the morphologically similar species. Incrimination of An. oswaldoi s.s. in malaria transmission in Brasil needs further investigation because other undescribed species from Acre may have been confounded with this taxon. Financial Support: FAPESP (Grant 05/53973-0); CNPq (Grant 472485/2006-7); UNICEF/UNDP/World Bank/WHO (TDR) (Grant A50252).

#### 215 Operational uses of mosquito systematics and taxonomy

Daniel Strickman, daniel.strickman@ars.usda.gov

Mosquito systematics is the study and description of phylogenetic and other relationships between taxa at all levels from family to below the species level. Taxonomy is a subset of systematics that applies the knowledge developed to establish systems for identification. Operational integrated mosquito management must find a balance between these disciplines to have useful taxonomy that reflects those aspects of systematics that produce biological insights with practical application. Species level changes have a history of improving operational applications, but superspecific changes based on systematics hypotheses have a more checkered history.

216 Evidence to support 3 conspecific cytological races of Anopheles campestris-like strains in Thailand Benjawan Tuetan, Visut Baimai, Atiporn Saeung, Sorawat Thongsahuan, and Wej Choochote, wchoocho@mail.med.cmu.ac.th

To verify the genetic proximity of *Anopheles campestris*-like strains from Chiang Mai, Maha Sarakham, Sa Kaeo, and Ayuttaya provinces, 6 isoline colonies of *An. campestris*-like forms [i.e.; Form B (X2, Y2) and E (X2, Y5): Chiang Mai; Form E (X2, Y5): Maha Sarakham; Form E (X1, X2, X3, Y5) and F (X2, Y6): Sa Kaeo; and Form F (X2, X3, Y6): Ayuttaya] were established in our insectary at Chaing Mai University. They were investigated for their genetic relationships by crossing tests and comparing the DNA sequences of rDNA internal transcribed spacer 2 (ITS2) and mitochondrial cytochrome c oxidase subunit I and II (COI and COII). The results of reciprocal and F1-hybrid crosses among 6 isoline colonies indicated that they were genetically compatible, providing viable progenies and completely synaptic polytene chromosomes. The sequences of ITS2, COI, and COII among 3 forms were nearly identical to each other (genetic distance < 0.005). Based on evidence of no post-mating reproductive isolations and nearly identical of DNA sequences of ITS2, COI, and COII regions among *An. campestris*-like Form B, E and F, we concluded that they are conspecific cytological races in Thai population.

#### 217 The Mosquito Barcode Initiative

Yvonne Linton and Richard Wilkerson, wilkersonr@si.edu

The Mosquito Barcode Initiative is an ongoing international collaboration based at The Natural History Museum in London and the Smithsonian's Laboratory of Analytical Biology near Washington, DC. The "barcode," which is now in widespread use as a means of characterizing "all" life, is a 750bp sequence from the mitochondrial COI gene. There has been excellent progress made by the MBI since an inceptive meeting 2 yr ago of mosquito researchers from all over the world, including a first ever inventory of the mosquito collections in London and Washington. We now know that there are about 85% of mosquito species available for barcoding in the 2 museums. In addition, sequencing has been carried out on about 16,000 fresh and archived specimens representing about 20% of mosquito species. This preliminary data has shown that the barcode discriminates most species, even those that are closely related. It is expected that the barcode sequence will greatly accelerate our ability to carry out identifications and find unrecognized taxa. Current support for the project is from the Consortium for the Barcode of Life and a group of private donors in London. Collaboration, and specimens, are solicited from all interested parties.

### 218 Introduction to DWFP activities at the USDA's Mosquito and Fly Research Unit Gary Clark, gary.clark@ars.usda.gov

This presentation will introduce researchers from the Mosquito and Fly Research Unit of the USDA's Agriculture Research Service who are involved in a unique variety of projects supporting the Department of Defense's DWFP Research Program.

#### 219 Efficacy of TalstarOne-treated vegetation for mosquito control

Daniel Kline, dan.kline@ars.usda.gov, Sandy Allan, David Dame, Max Meisch, and Todd Walker

Protection of deployed military personnel using conventional methods is often not practical. Objectives were: 1) assess perimeter barrier treatments in resting habitats, 2) develop small plot pesticide screening protocols, and 3) compare standard backpack versus electrostatic sprayers. Miniplots (10 ft x 10 ft) consisting of wax myrtles (Myrica cerifera) were established in FL and AR. Bifenthrin, (0.11 and 0.22 lb AI per acre) was applied. Efficacy was determined by trapping, landing counts, bioassays, and chemical residue analysis. Coleman MD-2500® mosquito traps were operated in mini-plots to collect mosquitoes. Bioassays were conducted with leaves from the plants in the mini-plots. Additional plants were used for collecting leaves for chemical residue analysis. Bioassays and residue analysis were conducted pretreatment, day of treatment, and weekly post-treatment until 24 h mortality was less than 10% in the bioassays. Gainesville cage studies used laboratory reared mosquitoes (Ochlerotatus taeniorhynchus and Aedes aegypti); in AR natural populations of Anopheles quadrimaculatus and Psorophora columbiae, produced in large numbers in rice-fields, were used. Bifenthrin was also applied (0.22 lb AI/acre) to perimeters (8 ft wide x 6 ft high) of field plots (0.1 acre) near rice-fields in Arkansas. Efficacy was determined in the same manner as for the mini-plots.

#### Sampling and analysis of pesticides from barrier treatments 220

Brian Quinn, brian.quinn@ars.usda.gov, Ulrich Bernier, and Daniel Kline

Barrier pesticide treatments on surfaces, including plants are one strategy to abate mosquitoes and other biting flies. Sampling and analysis of pesticides used in barrier treatments are important factors in determining the choices of application equipment, active ingredients, and frequency of treatments for a given location. The choice in sampling approach needs to be carefully considered to provide meaningful results and feedback with respect to qualitative and quantitative information about a particular barrier treatment. The quantitative results can provide information regarding application rate, uniformity of application over an area and the residual amount of active ingredient remaining post-application. Qualitative information is obtained from the analysis of samples and indicates potential breakdown products due to environmental factors. A technique such as gas chromatography/mass spectrometry (GC/MS) can play a vital role in the accurate assessment of residual pesticides in barrier treatments. This presentation will address the information that can be obtained from GC/MS analysis of barrier treatments, and appropriate sampling strategies to obtain useful results.

#### Discovery of novel biting fly repellents and insecticides from structure-activity studies

Ulrich Bernier, uli.bernier@ars.usda.gov, Alan Katritzky, Svetoslav Slovav, Gary Clark, and Kenneth Linthicum

The USDA historical archive of repellents and toxicants includes over 30,000 chemical structures that were identified and tested during the past 60 vr. We have undertaken a collaborative research project to target initially 6 subsets of these compounds to discover and develop new chemicals for personal protection and control of mosquitoes and biting flies. The first subset consisted of 68 candidate mosquito repellents and was comprised of piperidines and carboxamides. Evaluation of 2 dose rates using a cloth patch assay with human volunteers indicated that the duration of repellency from 1 of the carboxamides and 28 of the piperidines was greater than that of DEET at equivalent stoichiometric amounts. Some of the piperidines were repellent on cloth for over 50 d at the 25 µm dose, while DEET provided 7 d repellency using time to 5 bites (out of 500 mosquitoes) as the failure threshold. A recent study involving approximately 20 chemicals that are known or candidate residual insecticides for anopheline mosquitoes has been initiated. Recent progress made during this study will be reported.

#### A high throughput screening method to identify potential pesticides for mosquito control Julia Pridgeon, julia.pridgeon@ars.usda.gov, and James Becnel

Aedes aegypti (L.) is the primary vector of dengue and yellow fever both of which have become major international public health concerns. Insecticides remain a major component of integrated programs to control this medically important pest. To screen thousands of chemicals to identify potential pesticides for mosquito control, a high throughput screening method using 1<sup>st</sup> instar larvae of Ae. aegypti was created and evaluated in our laboratory. LD<sub>50</sub> values from a representative group of compounds were determined using this screening method and compared to LD<sub>50</sub> values determined with topical application against female adults of Ae. aegypti. Our results suggest that this high throughput screening method is suitable for screening thousands of chemicals quickly to identify effective chemicals.

#### Cytochrome c plays an important functional role in the development of Aedes aegypti

Liming Zhao, liming.zhao@ars.usda.gov, Julia Pridgeon, James Becnel, Gary Clark, and Kenneth Linthicum

Cytochrome c is a highly conserved protein that is found in many multicellular and unicellular organisms. Cytochrome c is a critical intermediate in apoptosis, a controlled form of cell death that kills cells as part of their natural process of development and in response to environmental condition. To detect whether cytochrome c in the mosquito Aedes aegypti (AeaCytC) is developmentally regulated, we employed quantitative real-time PCR to examine AeaCytC gene expression levels in different developmental stages of Ae. aegypti. Quantitative real-time PCR showed that AeaCytC was expressed in each larval and adult developmental stage and was highly expressed in teneral female Ae. aegypti. Aedes aegypti cytochrome c protein (AeaCYTC) was detected only in adult mosquitoes.

We also investigated the effect of certain environmental factors (temperature, ultraviolet light, and pesticides) on AeaCytC gene and AeaCyTC protein expression. Double-stranded RNA (dsRNA) synthesized from AeaCytC gene was injected into adult Ae. aegypti, resulting in high mortality. The present study suggests that AeaCytC plays an important functional role in the development of Ae. aegypti and may provide information useful for designing novel control strategies for mosquitoes and identifying new pathways to target for the development of molecular pesticides.

### 224 Behavioral definitions, 'excito-repellency', and its meaning with respect to mosquito contact with treated surfaces

Miriam Cooperband, miriam.cooperband@ars.usda.gov, and Sandra Allan

Pyrethroids are said to have 'excito-repellent' effects on mosquitoes. These effects were examined by studying facultative landing and resting behavior of mosquitoes when offered a choice between surfaces treated with 1 of 3 pyrethroids versus blank controls. Species from 3 genera were tested, *Aedes aegypti*, *Anopheles quadrimaculatus*, and *Culex quinquefasciatus*, and each species had its own distinctly different innate resting behavior that was differentially affected depending on the pyrethroid used. The results are discussed in the context of the terminology used to describe insect behaviors, with special attention to the term 'excito-repellency'.

### Overview of activities at the Navy Entomology Center of Excellence in Jacksonville, Florida Daniel Szumlas, daniel.szumlas@med.navy.mil, Todd Walker, Muhammad Farooq, and Craig Stoops

An overview of the command structure and function of the Navy Entomology Center of Excellence will be presented, highlighting its primary missions as well as various activities relating to the testing and evaluation of novel chemistries and application technologies for the Deployed Warfighter Protection Program.

#### 226 Structure-activity relationship studies on the arthropod repellent callicarpenal

Charles Cantrell, clcantrl@olemiss.edu, Jerome Klun, Julia Pridgeon, James Becnel, and Steve Duke

Based on botanical lore of insect repellent properties, essential oil extracts from the American beautyberry (*Callicarpa americana*) were recently investigated by bioassay-guided fractionation using the yellow fever mosquito, *Aedes aegypti*, leading to the isolation of callicarpenal (13,14,15,16-tetranor-3-cleroden-12-al). In previous studies, callicarpenal showed significant bite-deterring activity against *Ae. aegypti* and *Anopheles stephensi*, in addition to repellent activity against host-seeking nymphs of the blacklegged tick, *Ixodes scapularis*. In the present study, structural modifications were performed on callicarpenal in an effort to understand the functional groups necessary for maintaining and/or increasing the activity. Among the modifications that will be discussed are the oxidation and reduction of the C-12 aldehyde to its corresponding acid or alcohol, complete reduction of the C-3 olefin, epoxidation of the C-3 olefin, and various methyl ester and acetate products. Combinations of the above modifications will also be discussed as well as their effects on both biting-deterrency and toxicity against *Ae. aegypti*.

### Plant derived compounds and their synthetic analogs against the yellow fever mosquito Aedes aegypti (Diptera: Culicidae)

Kumudini Meepagala, kmeepaga@olemiss.edu, Julia Pridgeon, George Sturtz, James Becnel, and Stephen Duke

Aedes aegypti (L.) is considered the primary vector for both dengue and yellow fever. Application of insecticides is one of the major control methods for this medically important insect pest. However, few new insecticides have been developed for mosquito control. As part of a collaborative effort to search for new insecticides to control mosquitoes, natural products isolated from various plant families and some synthetic derivatives of piperidines and amides were evaluated against adult Ae. aegypti. Natural products offer diverse classes of compounds with varying modes of action. In laboratory bioassays based on 24-h LD<sub>50</sub> values after topical application, the most toxic compound among the piperidines was 2-ethyl-1-undec-10-enoyl-piperidine (LD<sub>50</sub> = 0.80  $\mu$ g per mosquito). Piperine [(E, E)-1-piperoyl-piperidine], isolated from Piper nigrum, was less toxic (LD<sub>50</sub> = 8.13  $\mu$ g per mosquito) than all the tested ethyl- or methyl- derivatives of 1-undec-10-enoyl-piperidines. Vulgarone and apiol, isolated from Artemesia douglasiana and Ligusticum hultenii, had LD<sub>50</sub> values of 1.5 and 5  $\mu$ g per mosquito respectively. Synthesis, isolation, and toxicity evaluation of these compounds will be discussed.

#### 228 Prospective use of fast-acting pyrethroids for vector control

Kamlesh Chauhan, kamal.chauhan@ars.usda.gov, Thanh Lee, and John Grieco

Fast acting pyrethroid insecticides like transfluthrin and metofluthrin are used in household and hygiene products, mainly for the control of flying insects such as mosquitoes and flies. Transfluthrin has been registered for use in about 50 countries worldwide, while metofluthrin is recently registered in few. The main formulation types available are mosquito coils and aerosols. High vapor pressure is the main physiochemical property of these and related fast-acting pyrethroids. In our study, we evaluated molecular properties of such pyrethroids for efficacy, commercial viability, as well as adaptation to new long-lasting formulations. An interesting feature revealed for these candidate pyrethroids was susceptibility to numerous resistant species of mosquitoes.

#### 229 Development of spray system for UAV's

Clint Hoffmann, choffmann@tamu.edu, Yanbo Huang, and Bradley Fritz

This presentation will summarize the current state of development and testing of an automated spray system that has been designed to fit on an autonomous unmanned aerial vehicle (UAV). The spray system has been designed to deliver spray materials that can be used in vector control scenarios.

#### 230 Natural pesticides and repellents from plant essential oils

Marc Dolan, Janet McAllister, jvm6@cdc.gov, Nicholas Panella, Gabrielle Dietrich, Joseph Piesman, and Joseph Karchesy

The biocidal activities of essential oils and their components extracted from Alaska yellow cedar (*Chamaecyparis nootkatensis*) were evaluated against adult mosquitoes (*Aedes aegypti*), fleas (*Xenopsylla cheopis*), and nymphal ticks (*Ixodes scapularis*). Laboratory bioassays were conducted to determine the in vitro dose-mortality data through 24 h. In addition, we demonstrated the laboratory efficacy of these natural products as both acaricides and repellents against the ticks that transmit Lyme disease spirochetes. Limited field trials have been initiated in a Lyme endemic region in collaboration with Oregon State University and the Freehold Township Health Department, NJ, using 2 different formulations of naturally derived products that demonstrated significant levels of control for up to three weeks against field populations of Lyme disease spirochete infected ticks.

#### 231 Pyriproxyfen as a vector control tool

Gregor Devine, Amy Morrison, Helvio Astete, Victor Lopez, and David Florin, david.florin@med.navy.mil

Field comparisons of larvicides around the Amazon city of Iquitos, Peru, show that pyriproxyfen is an impressive tool for the control of mosquito disease vectors (*Aedes*, *Culex*, and *Anopheles* spp). We present the results of large trials (>1500 households) comparing different larvicides (pyriproxyfen, temephos) and treatment strategies against *Ae. aegypti*. Ecotoxicological data from large water bodies (disused fishponds) shows that even at very low doses (50 ppb) pyriproxyfen caused dramatic decreases in the emergence of a variety of mosquito species but had relatively minor impacts on sensitive environments. Data from adult emergence traps and sieved water samples showed that treatment with pyriproxyfen did not affect the abundance or development of most non-target arthropods (e.g. Odonata, Hemiptera, Coleoptera, Trichoptera, and Ephemeroptera). We also present field data showing that adult female mosquitoes resting on surfaces dusted with pyriproxyfen can transfer enough larvicide to other mosquito sites to affect immature mosquitoes developing therein. This phenomenon may have considerable potential for development as a novel control tool. We conclude that pyriproxyfen is the most effective larvicide currently available, that its optimal use can make a real contribution to mosquito control, and that it can be used in the wider environment without undue risk.

#### 232 Product development made simple: A quarter billion dollars and 10 years

John Paige III, john.paige@bayercropscience.com, and Peter Connelly

Product development and registration in the USA has become increasingly complex, time consuming, and costly. Required studies and regulatory reviews now require ca. 10 yr from discovery to registration, and on average, companies screen over 200,000 new compounds per yr in order to discover 1 or 2 new products. This time and effort require significant investment. In 2004, the 10 major worldwide chemical companies spent, on average, ca. \$240 million dollars for each product brought to market. Because of this complexity and cost, only the strong survive. Industry consolidation of basic manufacturers has occurred significantly since 1990 in all areas, including the public health sector. Consolidation increases critical mass and experience, which can help speed innovation. An example of such innovation is the collaboration between Bayer and the Bill and Melinda Gates Foundation to combat malaria with better mosquito control products.

#### 233 How safe is safe enough? Mosquito adulticides and public health risks

Robert K. D. Peterson, bpeterson@montana.edu, Jerome J. Schleier III, and Ryan S. Davis

The emergence of West Nile virus in North America in 1999 and the USEPA re-registration process for mosquito adulticides have turned public attention to health and ecological risks associated with the use of insecticides for vector management. Several risk assessments and allied biomonitoring and epidemiology studies have been conducted to date. All show that the human health risks from adulticiding are negligible. The risk assessments that have been conducted represent reasonable worst-case insecticide exposure estimates that were derived from generic environmental fate models. Data on actual environmental concentrations of adulticides have been lacking. These data are needed to determine the conservatism of the risk assessments that have been conducted and to refine existing risk assessments. We review the existing weight-of-evidence on risks associated with adulticide use and present our recent environmental insecticide fate data to show that measured insecticide concentrations result in lower exposures, and therefore lower risks, than those originally estimated in reasonable worst-case risk assessments.

#### 234 Mosquito control in FL—an overview

James Clauson, clausoj@doacs.state.fl.us

FL law, Chapter 388 of the FL Statutes, regulates mosquito control in FL. The Department of Agriculture and Consumer Services is responsible for enforcing and regulating this law. The Bureau of Entomology and Pest Control has a mosquito control section that oversees the mosquito control districts in FL. If a district wishes to be a state-approved program and receive state grant monies, they must meet certain requirements and submit monthly accomplishment reports to the mosquito control section office. Currently, there are 60 state-approved districts, which have a total combined budget of approximately \$142,000,000.

#### 235 Developing community outreach partners

David Zazra, dzazra@nsmad.com

In 2006, the North Shore Mosquito Abatement District and the North Shore Senior Center began to develop a program to encourage people to protect themselves from mosquito-borne illnesses. With the northeastern area of IL a hotbed of West Nile virus activity, the project became focused on preventing WNV. Volunteers from the North Shore Senior Center received training from the NSMAD in repellent usage, prevention tips, basic scientific information, statistics, etc. These volunteers then went out into their communities to help spread the word through various public forums. Farmer's markets, senior care facilities, sidewalk sales, summer festivals, and block parties were just a few of the locations where volunteers spoke about the dangers of WNV.

#### 236 Scouting and mosquito control: Partners in education

Pamela C. Jacobson, JacobsonP@hillsboroughcounty.org, and Michael F. Muench

As part of our community outreach effort, Hillsborough County Mosquito and Aquatic Weed Control developed the Mosquito Control Patrol Scouting Activity Patch Program in June of 2007 to encourage boys and girls who belong to scouting organizations to learn about mosquito control and earn a colorful activity patch. The program features a PowerPoint presentation, sample 1-d event schedule, various games and educational activities, patch design and ordering information, and other supporting documents and instructions. We have packaged it all on a CD, and will provide copies free of charge for other mosquito control organizations wanting to develop their own scouting activity programs.

#### 237 Revamp your educational material

Carl I. Sivertsen, carl.sivertsen@fairfaxcounty.gov, and Jorge Arias

Since the first signs of West Nile virus, efforts have been made to keep the community well informed of personal protection and mosquito control measures that needed to be implemented. Informational material was prepared and distributed across the community for 4 yr. Then in late 2005 and during the 2006 mosquito season, the information campaign stalled. At community events, residents passed up the information by politely saying "No thanks, I already have one." Were we victims of our own success? How could we get the information back in the hands of the public? Normally, residents repeatedly and eagerly accepted or requested our educational materials. To get the message out, we took steps to re-package our information to regain community interest and a want for 'new information'. The new look and added information has resulted in renewed interest from residents and once again we heard "Thank you very much, this information is very helpful." Effective community education and outreach is a critical component to the success of a mosquito control program. By bringing and keeping the community involved with mosquito control, residents learn how mosquito control protects the public's health and enhances quality of life.

#### 238 Developing a service request application for mosquito control - Case study

Suzanne Luinis, tnmapping@comcast.net

During the mosquito season in the Northeast it is common to enter a mosquito control office and hear the constant ringing of phone and fax lines - citizens requesting service and relief from mosquitoes. Receiving, logging, and servicing the thousands of requests has become a challenge for many of the districts. This presentation will focus on the various components of developing a service request application for mosquito control. From data entry forms to log requests, to the use of PDAs to record completed service request information, all phases of the service request life cycle will be discussed and technology introduced to respond to these requests in a timely/cost-effective fashion. True North Mapping, Inc. is a GIS/GPS consulting firm, specializing in custom mobile application development. During the presentation, True North will demonstrate a service request application developed for Bristol County Mosquito Control Project, MA.

#### 239 Area-wide management of the Asian tiger mosquito

Dina M. Fonseca, dinafons@rci.rutgers.edu, Gary Clark, Randy Gaugler, and Daniel A. Strickman

The Asian tiger mosquito, *Aedes albopictus*, is often responsible for most complaints to urban mosquito control programs, yet there is no clearly defined protocol for controlling this species. We are developing a multidisciplinary approach grounded by an economic analysis, which incorporates extensive public education and involvement, with focused application of established biological and chemical control interventions. Our methodology will be developed in 2 NJ counties. In Phase I, we will perform an initial assessment of *Ae. albopictus* abundance, dispersal, population structure, and insecticide resistance status, as well as of its social and economic

impact. In Phase II, we will compare *Ae. albopictus* abundance and associated human behavior in 2 pairs of experimental and control plots of 1,000 homes (up to 640 acres) each. In Phase III, we will expand our control efforts county-wide (>140,000 acres) initially targeting source populations. Throughout the project, the costs of all control procedures will be documented and an economic analysis will be conducted. If we succeed in controlling the Asian tiger mosquito and our strategy is economically sound, we will proceed to Phase IV and expand the initiative nationally by recruiting 12 geographically diverse mosquito control agencies to implement our methodology.

### 240 Identification of *Culex* species using species-specific PCR primers and its application in WNV surveillance program

Mahmood Iranpour, iranpour@cc.umanitoba.ca, Robbin Lindsay, and Antonia Dibernardo

In 2002, more than 17,000 mosquito pools collected in Canada (Quebec, Ontario and Manitoba), were tested at the National Microbiology Laboratory in Winnipeg, Manitoba for infection with West Nile virus (WNV). Using real-time RT-PCR, 558 mosquito pools (86% *Culex* species and 14% other species) had evidence of infection with WNV. However, only 30% of the *Culex* specimens were identified to the species level. In this study, *Culex* species-specific PCR primers were designed to identify individual mosquitoes and mixed pools of *Culex* mosquitoes to species. In addition, pools of non-*Culex* mosquitoes that tested positive for WNV were also screened for *Culex* DNA to determine the frequency of cross contamination among mosquitoes of different species. DNA extracts from 121 *Culex* and 51 non-*Culex* pools, previously positive for WNV, were screened and *Culex* DNA was detected in approximately 6% of non-*Culex* pools. This study demonstrates that contamination among mosquito species can occur and emphasizes that precautions should be taken to minimize this potentially confounding effect.

### 241 Efficacy of mosquito control strategies and potential for non-target effects in Seattle, WA Morgan R. Sternberg, seastern@u.washington.edu, C. G. Grue, J. M. Grassley, K. A. King, and L. L. Conquest

In anticipation of the spread of West Nile virus, Seattle officials promoted a city-wide research effort spanning the summers of 2006 and 2007 designed to investigate the efficacy and fate of 4 common larvicides: Mosquito Dunks® and Bits® (AI = Bti), VectoLex® WSP (AI = B. sphaericus), VectoLex® CG (AI = B. sphaericus) and Altosid® Briquets (AI = methoprene). All treatments resulted in a rapid reduction in number of pupae (Bti and B. sphaericus treatments) or emergence success of pupae (methoprene). The B. sphaericus treatment was the most efficacious in 2006. Because of these results, VectoLex CG was chosen for city-wide application in 2007. Efficacy was realized for 7 wk; in most cases though, a relatively high number of precipitation events confounded the direct effect of the larvicide. The larvicide was detected above background levels at least 1 wk post-treatment in each of the basins chosen for monitoring larvicide fate; subsequent precipitation events likely decreased the B. sphaericus concentrations. The larvicide was detected in 2 particular sets of basins prior to application suggesting input from outside sources. Larvicide was detected in 3 urban creeks connected to treated basins. Preliminary results from toxicity tests suggest that concentrations of B. sphaericus and Bti detected in each of the watersheds pose little hazard to juvenile salmonids.

#### New mosquito biolarvicide formulation for improved residual activity

Margaret E. Lyn, Margaret.Lyn@ARS.USDA.GOV, Douglas Streett, and James Becnel

Novel biolarvicide formulations were designed and developed to enhance residual activity of *Bacillus thuringiensis* Berliner var. *israelensis* (*Bti*). These formulations were developed specifically to maintain the active ingredient in the upper feeding zone and to provide efficacy regardless of water quality.

#### **Poster Session Abstracts**

#### P-01 Effects of wind speed on aerosol spray penetration in adult mosquito bioassay cages

W. Clint Hoffmann, choffmann@tamu.edu, Brad Fritz, Muhammad Farooq, and Miriam F. Cooperband

Bioassay cages are commonly used to assess efficacy of insecticides against adult mosquitoes in the field. To properly correlate adult mortality readings to insecticidal efficacy and/or spray application parameters, it is important to know how the cage used in the bioassay interacts with the spray cloud containing the applied insecticide. This study compared the size of droplets, wind speed, and amount of spray material penetrating cages and outside of cages in a wind tunnel at different wind speeds. Two bioassay cages, CMAVE (Center for Medical, Agricultural and Veterinary Entomology) and Circle, were evaluated. The screen materials used on these cages reduced the size of droplets, wind speed, and amount of spray material inside the cages as compared to the spray cloud and wind velocity outside of the cages. When the wind speed in the dispersion tunnel was set at 0.6 m/sec (1.3 mph), the mean wind speed inside of the CMAVE Bioassay Cage and Circle Cage was 0.045 m/sec (0.10 mph) and 0.075 m/sec (0.17 mph), respectively. At air velocities of 2.2 m/sec (4.9 mph) in the dispersion tunnel, the mean wind speed inside of the CMAVE Bioassay Cage and Circle Cage was 0.83 m/sec (1.86 mph) and 0.71 m/sec (1.59 mph), respectively. Consequently, there was a consistent 50-70% reduction of spray material penetrating the cages compared to the spray cloud that approached the cages. These results provide a better understanding of the impact of wind speed, cage design and construction on ultra low volume spray droplets.

### P-02 Evaluation of passive and active control measures against mosquito vectors in western Thailand Brian P. Evans, Brian.Evans@afrims.org, and Jason R. Richardson

In the foreseeable future, a large contingent of US forces will remain deployed in southeast and southwest Asia, where mosquito-borne disease (i.e., malaria) is endemic and has the capability to compromise military operations. Field research is currently underway in western Thailand to determine the efficacy of select passive (i.e. insecticide-impregnated nets and tents) and active (i.e., barrier spray) control measures that would effectively minimize the threat of mosquito-borne disease to the health of deployed soldiers. Several parameters are being used to measure the efficacy of control. These include mosquito human landing counts, parity rates, and rates of *Plasmodium* infection among collected mosquitoes.

### P-03 Laboratory evaluation of Callicarpenal americana essential oil for toxicity against female adults of Aedes aegypti

Julia W. Pridgeon, julia.pridgeon@ars.usda.gov, Charles Cantrell, and James J. Becnel

As part of a collaborative effort to search for new insecticides to control mosquitoes, essential oil from the American beautyberry, *Callicarpenal americana*, was evaluated in laboratory bioassays using topical application against female adults of *Aedes aegypti*. *Callicarpenal americana* oil had an LD<sub>50</sub> value of 9.91  $\mu$ g per mosquito (24-h mortality), which is slightly higher than the LD<sub>50</sub> value of piperine (8.13  $\mu$ g per mosquito), which is a piperidine alkaloid derived from *Piper longum* known to have insecticidal activity against mosquitoes. The toxicities of 3 chemicals isolated from the *Callicarpenal americana* essential oil were also evaluated. The most toxic chemical evaluated was intermedeol, with an LD<sub>50</sub> value of 4.66  $\mu$ g per mosquito. The second most toxic chemical was callicarpenal, with an LD<sub>50</sub> value of 5.13  $\mu$ g per mosquito. Humulene epoxide II, the third chemical ingredient from the *Callicarpenal americana* essential oil, showed the lowest activity, with an LD<sub>50</sub> value of 8.85  $\mu$ g per mosquito. Taken together, this preliminary data suggest that intermedeol and callicarpenal might be suitable templates for structure-activity analysis for the design of new insecticides for mosquito control.

### P-04 Post-mission weather analysis of aerial applications using FLIGHTMASTER and AIMMS 20 equipped aircraft

Robert Mickle, Bob.Mickle@REMSpC.com, and William Jany

The use of on-board meteorological instrumentation and second-generation aerial guidance systems allows for post-mission analysis at a level never before reported. This poster reviews examples of several missions that give a good example of the meteorology experienced during routine and emergency spray missions throughout the US. The interpretation of this data can help in mission planning for future applications.

### P-05 Host species diversity and post-blood feeding carbohydrate availability enhance survival of females and fecundity in *Aedes albopictus* (Diptera: Culicidae)

Arshad Ali, umar@ufl.edu, Rui-De Xue, and Donald R. Barnard

Adult survival of caged female *Aedes albopictus* that were fed on blood of 2 different hosts (double meal) was higher than the females fed only on 1 host (single meal) (mean survival:  $70.2 \pm 9.6$  vs.  $55.5 \pm 5.5$  %, respectively) when held in the laboratory for 72 h after blood feeding. Mean survival of females provided 10% sucrose solution (in water) after a single or double blood meal was higher ( $90.5 \pm 6.4$  % and  $89.3 \pm 6.5$  %, respectively) than in the respective groups receiving water only following blood feeding (double meal:  $49.0 \pm 9.6$  %; single meal: and  $45.3 \pm 10.9$  %). Females receiving a double meal were more fecund on average (89.0 + 6.6 eggs) than females provided a single meal ( $82.3 \pm 8.2$  eggs).

#### P-06 Mosquito bionomics in Sabine National Wildlife Refuge, LA: Nocturnal periodicity

William Dees, wdees@mcneese.edu, Linda Canning, Harry Meyer, Josiah Land, Caroline Chapman, Mukesh Wagle, Benjamin Clark, Joseph Dees, and Terry Sylvester

A nocturnal periodicity study of mosquitoes during new moon phases in the Sabine National Wildlife Refuge in southwestern LA is underway. Mosquitoes are collected before, during and after sunset and sunrise, and at other intervals throughout the night using a modified Centers for Disease Control mosquito light trap with a rotating collector placed 1.5 m above ground. This trap is placed in an area with little to no competing light and where no pesticide applications are conducted. Studies were initiated in July 2006. To date, *Aedes sollicitans*, *Anopheles crucians* and *Culex salinarius* have been the most commonly collected species. Data from this study will broaden our understanding of mosquito population dynamics in southwest LA. Studies such as this are excellent for undergraduate research projects.

#### P-07 Mosquito bionomics in Sabine National Wildlife Refuge, LA: Nocturnal vertical distribution

William Dees, wdees@mcneese.edu, Linda Canning, Harry Meyer, Josiah Land, Caroline Chapman, and Mukesh Wagle

A vertical distribution study of mosquitoes in the Sabine National Wildlife Refuge in southwestern LA was conducted in fall 2006 and spring 2007. This study provides information on nuisance mosquitoes and disease vector ecology. Mosquitoes were collected using Centers for Disease Control mosquito light traps placed overnight at 1.5 m, 3.5 m, and 5.5 m above ground. Traps were placed in areas with little to no competing light and where no pesticide applications are conducted. Meteorological conditions were recorded. *Anopheles crucians* was the most

common species at 1.5 m. *Culex salinarius* was the most common species at 5.5 m. Information from this study will broaden our understanding of mosquito population dynamics in southwest Louisiana. Studies such as this are excellent for undergraduate research projects.

### P-08 Evaluation of mosquitocidal activity of *Litsea elliptica* Blume (Lauraceae) leaf extract against the dengue vector *Aedes aegypti Linnaeus* (Diptera: Culicidae)

Hidayatulfathi Othman, hida@medic.ukm.my, Sallehudin Sulaiman, Ibrahim Jantan, and Zaridah Mohd. Zaki

Growing awareness in using ecofriendly and biologically compatible phytoconstituents as natural insecticides and repellents for the safety of life and ecological balance led to conscientious efforts by scientists all over the world to search for alternative sources of plant derivatives for effective use as mosquitocides. Encouraged by this, Litsea elliptica Bl. leaf extract viz: hexane fraction, chloroform fraction, ethyl acetate fraction and methanol fraction and essential oil were investigated for anti-mosquito potential including larvicidal, adulticidal, ovicidal, and repellent activities against Aedes aegypti, the vector of dengue haemorrhagic fever. These fractions and essential oils were screened for larvicidal activity on early 4th instar larvae of Ae. aegypti following WHO standard protocol. The hexane fraction was found to show the highest larvicidal effect with the median lethal concentration (LC<sub>50</sub>) value of 1.88 ppm and the LC<sub>90</sub> value of 10.76 ppm, respectively. Ovicidal activity was also demonstrated using the hexane fraction against the eggs of Ae. aegypti with LC<sub>50</sub> value at 39.46 ppm and the LC<sub>90</sub> value at 269 ppm. The mortality of the newly hatched larvae, reared from eggs in this media, reached 100% at concentrations as low as 100 ppm, 2 days post-hatching. However, when tested for the adulticidal activity, the essential oil showed promising adulticidal effect with the  $LC_{50}$  and  $LC_{90}$  values of 0.04 and 0.90 mgcm-2 respectively. It was then further tested for biting deterrence in human volunteers. In laboratory, L. elliptica essential oil possessed repellent effect against Ae. aegypti with ED<sub>50</sub> value of 0.0003 mgcm-2. Topical application of the essential oil did not induce dermal irritation. Litsea elliptica, therefore, can be considered as a probable source of some biologically active compounds used in the development of mosquito control agents.

### P-09 Evaluation of four trap types using the BG-lure in mosquito surveillance in Fairfax County, VA Hina Bhalala, hina.bhalala@fairfaxcounty.gov, and Jorge Arias

Since the detection of West Nile virus (WNV) in Fairfax County in 2000, an intensive mosquito surveillance program including WNV laboratory testing was established. With a variety of WNV bridge vector species in the area, it is essential to use a more efficient trap to collect the maximum number of these mosquito species in order to test for WNV and provide a more accurate estimation of the true burden of species in this area. In a previous study, we illustrated the increased efficiency of the BG-Sentinel trap in the collection of *Aedes albopictus* relative to the industry standard,  $CO_2$ -baited CDC light traps. In a transitional habitat, this study used 4 replicates of a 4x4 Latin square to evaluate the efficacy of four trap types baited with the BG-lure: BG-Sentinel, Faye-Prince, CDC light, and the Zumba trap. The results of this study evaluate the efficiency and effectiveness of 4 trap types baited with  $CO_2$  and the BG-lure in the collection of WNV vector species. In addition, the value of incorporating the vector index as a surveillance tool and the BG-lure bait into the routine mosquito surveillance in Fairfax County was also assessed.

#### P-10 Feeding preferences of vectors of eastern equine encephalitis in TN

Sara B. Cohen, sara.cohen@state.tn.us, Kenneth Lewoczko, Dora B. Huddleston, Sudeshna Mukherjee, Timothy F. Jones, and Abelardo C. Moncayo

Traditional transmission of eastern equine encephalitis (EEE) occurs in coastal regions by the enzootic mosquito *Culiseta melanura*. Transmission in TN, an inland region, takes place around wetlands where the predominant vectors are *Culex erraticus*, *Cx.restuans*, *Anopheles quadrimaculatus*, *Coquillettidia perturbans*, and *Aedes vexans*. In order to assess the vectorial capacity of other mosquito species for virus transmission, we collected blood fed mosquitoes from resting boxes set around a swampland that was at the epicenter of an equine outbreak of EEE in 2005. Traps were set and aspirated from June through September of 2006. 12,459 female and 15,239 male mosquitoes were collected and 2,817 females were found to be blood fed. We identified the blood meal sources from *Cx. erraticus*, *Cx. restuans*, *An. quadrimaculatus*, and *An. punctipennis* as avian or mammalian by using PCR to amplify a portion of the cytocrhome b gene. Out of 29 *An. quadrimaculatus* tested, 12 (41%) showed an affinity for avian blood, 23 (79%) for mammalian blood, and 6 (20%) had both blood meal types. Preliminary results suggest that *An. quadrimaculatus* is a likely bridge vector for arbovirus transmission to humans. Data on other species as well as sequencing results of the amplicons will be presented.

### P-11 Nighttime temperatures and the onset of WNV in West Valley, San Bernardino County, CA T. Steven Su, tsu@wvmvcd.org, and Min-Lee Cheng

West Nile virus was first detected in a dead crow in the southwestern corner of San Bernardino County, CA in 2003. In the ensuing year, a WNV outbreak occurred in the region, involving a score of positive mosquito samples, dead birds, and sentinel chickens in the early season, followed by detection of human and equine cases in mid-July and early August respectively. A similar, but less aggressive, WNV infestation was recorded in 2005. In 2006 and 2007, even fewer positive mosquitoes, dead birds, and sentinel chickens were detected and no human and equine cases were reported. Available surveillance data on WNV detection in mosquitoes, dead birds, sentinel chickens, equines, and humans for 4 consecutive yr suggest that the onset of WNV infections was postponed for 4 wk in

2005, for 10 wk in 2006, and for 12 wk in 2007 in comparison to 2004. These delays in onset seem to correlate to the lower nighttime temperatures in spring and/or early summer in 2005, 2006 and 2007.

### P-12 Temporal and spatial distribution of mosquito species at Nisqually National Wildlife Refuge, WA Elizabeth Dykstra, elizabeth.dykstra@doh.wa.gov, Shannon Davis, Sonya Ramos, and JoMarie Brauner

Mosquitoes were collected weekly from May 9 through September 27, 2007 using 2 EVS light traps baited with dry ice. One trap was placed in a wooded area and the second trap was placed close to an open field and adjacent to a pond. The traps were approximately 200 m apart and were set from 4:00 PM to 7:00 AM. Upon collection, mosquitoes killed by freezing and then sorted according to species. Species collected in the greatest numbers included *Coquillettidia perturbans*, *Culex pipiens*, *Cx. tarsalis*, *Anopheles punctipennis*, *Culiseta particeps*, *Ochlerotatus sierrensis*, *Oc. aboriginis*, *Oc. increpitus*, and *Oc. dorsalis*. *Coquillettidia perturbans* was the most common species collected at both sites, followed by *Cx. tarsalis* at the pond site and *Oc. aboriginis* at the wooded site. Any *Cx. pipiens* and *Cx. tarsalis* caught in quantities of 10 or more were pooled and submitted for testing for West Nile virus (WNV) at the US Army's CHPPM-West, Ft. Lewis, WA. All pools tested negative for WNV. These data provide information on mosquito population dynamics in a western WA marsh and riverine environment.

# P-13 Temporality and diversity of mosquito species in urban and suburban King County, WA Sharon G. Hopkins, sharon.hopkins@kingcounty.gov, Natasha Close, Elizabeth A. Dykstra, Leah Helms, Eric Coker, and Peter Isaksen

In October of 2006, West Nile virus (WNV) made its first appearance in King County (KC). As a result, Public Health - Seattle & King County, in collaboration with the City of Seattle, intensified mosquito surveillance in 2007. EVS traps with dry ice were placed in diverse settings throughout KC each week. Mosquitoes were sorted by species by the WA Department of Health and pools of *Culex pipiens* were tested for WNV using PCR. In 2007, over 800 pools were collected and 200+ tested (all negative). *Culex pipiens*, *Ochlerotatus increpitus*, and *Coquillettidia perturbans* were the principal species until mid-July when *Cx. pipiens* and *Culiseta incidens* became the primary species. Mosquitoes were the most abundant within urban Seattle, with *Cx. pipiens* being the predominant species. The second most frequently-identified urban species was the nuisance mosquito, *Cs. incidens*. An urban amusement park in central Seattle consistently produced the largest number of mosquitoes, which were primarily *Cx. pipiens*. In contrast, collections from suburban or rural settings were more diverse with proportionally few *Cx. pipiens*. With the exception of a site in south KC where a large number of *Oc. increpitus* were collected, mosquito abundance was lower outside of urban Seattle.

### P-14 Importance to molecular diagnostics of extensive sequence diversity in insecticide resistance genes in mosquito vectors

William G. Brogdon, wgb1@cdc.gov, Ling Zhou, Tara Brant, and Robert A. Wirtz

Detoxification enzyme and target site resistance genes in vector species are shown to exhibit extensive sequence diversity within natural populations. Sequence data are presented for esterase, oxidase, and kdr resistance genes in *anopheline* and *culicine* species including *Anopheles gambiae*, *Aedes aegypti*, and *Culex* species. The importance of careful attention to this diversity in the design of diagnostic molecular probes is discussed. Also discussed are the opportunities this diversity presents for developing a deeper understanding of vector population biology, particularly as it reflects the exposure history of mosquito populations to control activities.

### P-15 CalSurv Gateway: Large-scale data management for vector and virus surveillance in CA Bborie K. Park, bkpark@ucdavis.edu, Bruce F. Eldridge, Christopher M. Barker, and William K. Reisen

Historically, mosquito abatement agencies have recorded surveillance data on paper or in short-term electronic formats such as spreadsheets. Relational databases, the proliferation of the Internet, and new web development methods now permit centralized data management. In CA, we have developed a framework for data collection, report generation, and visualization, which now serves vector control and public health agencies statewide. Laboratory test results are reported in real-time and all data are available in a variety of formats including trends for the current year plotted against previous averages, mapped surveillance results, and calculated mosquito infection rates. Aggregating data electronically permits not only analyses for current data, but permits the development of multi-year datasets for retrospective analyses.

#### P-16 Withdrawn

### P-17 Riceland mosquito management practices for Anopheles quadrimaculatus R.A. Allen, rallen@uark.edu, C.N. Lewis, W.W. Wilkes, and M.V. Meisch

Rice field surveillance of immature *Anopheles quadrimaculatus* Say mosquitoes was conducted in AR and MS to determine the effects of lambda-cyhalothrin applied for rice water weevil on natural mosquito populations. Sampling was conducted weekly. Lamda-cyhalothrin had no apparent effect on the population. An evaluation of aerial ULV larviciding was conducted using undiluted liquid *Bti* against *An. quadrimaculatus* in mid and late-season tall rice cultivars with control nearing 100%.

#### P-18 A 3-year analysis of RAMP and RT-PCR testing for WNV detection at the county level

Eric Williges, ewilliges@mercercounty.org, and Ary Farajollahi

We examine the value of the rapid analyte measurement platform (RAMP) and its comparison to the RT-PCR test for West Nile virus activity in Mercer County, analyzing data from 2005 to 2007. We also will examine and analyze the practical and scientific benefits and concerns of each system and ways they are used within our county.

### P-19 The gravid trap - Its place in an integrated mosquito surveillance and control program Fran Krenick, frankrenick@clarkemosquito.com

As a result of continued increases in human and animal illness across the country caused by mosquito-borne viruses, the importance of establishing and maintaining sensitive surveillance within mosquito control programs becomes critical. This poster will focus on one aspect of a sensitive surveillance network, the collection of already blood-fed female mosquitoes that are actively seeking oviposition sites. As adulticide applications increase with the detection of virus or disease in a particular area, the effectiveness of those control efforts can be determined by several methods. This poster will discuss the relationship of the gravid trap and the interpretation of the data collected as it relates to minimum infection rates in the field and overall mosquito populations.

#### P-20 Comparative evaluation of several mosquito trap configurations

John P. Smith, john.smith@famu.edu, Jimmy D. Walsh, and Eric H. Cope

Several commercial trap configurations were contrasted in Latin-square experiments to determine the number and species caught. Comparisons were made with different infusion water mixtures and power sources. Results will be presented in statistical and graphic form.

### P-21 Sequence and secondary structure of the rDNA ITS2 in North American Culicinae mosquitoes Brian D. Byrd, bbyrd@tulane.edu, and Dawn M. Wesson

The utility of diagnostic PCR-based assays to distinguish morphologically similar and cryptic mosquito species is well established. Likewise, the value of DNA sequences for systematic and phylogenetic studies is also clear. Today these technologies are routinely used in many laboratories and the second internal transcribed spacer (ITS2) region of ribosomal DNA is an often-used molecular marker for mosquito studies. Here we demonstrate the utility of ITS2 for phylogenetic analyses of North American Culicinae mosquitoes at different taxonomic levels with novel and previously reported sequences. Furthermore, we review the use of ITS2 in the Culicinae literature and consider the levels of reported intra- and interspecific sequence variation, the role of secondary structure in analyses, the potential impact of these observations on taxonomic/phylogenetic studies, and recommendations for future studies.

# P-22 Advances in a new long lasting impregnated mosquito net (LLIN) technology for malaria control - DuraNet® - provide improved protection against malaria transmission and lowers distribution burdens Rod Flinn, rodflinn@clarkemosquito.com

This poster will focus on a new long lasting impregnated mosquito net (LLIN) called DuraNet® that uses a specially controlled release system to deliver longer and more economical protection in combating malaria.

#### P-23 Multiple genes expressed in response to heat shock stress of Aedes aegypti adults

Liming Zhao, liming.zhao@ars.usda.gov, Julia W. Pridgeon, James J. Becnel, Gary G. Clark, and Kenneth J. Linthicum

Temperature is important for mosquito development and physiological response. Several genes of heat shock protein (HSP) families are known to be expressed in mosquitoes and may be crucial in responding to stress induced by elevated temperature. To determine other genes expressed in response to heat stress, we combined 2 powerful technologies, PCR-based cDNA subtraction and RNA interference (RNAi). PCR-select cDNA subtraction was used to identify target transcripts to heat shock treatment in adult *Aedes aegypti*. Heat shock treatment of adult *Ae. aegypti* was carried out for 1 h at 42 °C. Target transcripts up-regulated by heat shock included some HSPs as well as many other genes, such as cytochrome c oxidase, serine-type enodpeptidase, and glutamyl aminopeptidase. Additionally, 3 unknown protein up-regulated gene sequences were obtained. Up-regulated transcripts from heat shock treatment were further confirmed by real-time PCR. RNAi, using several genes identified as being up-regulated by heat shock treatment were injected into adults *Ae. aegypti* resulting in high mortality. High temperatures can drastically alter the gene expression of a vector mosquito population; understanding this process at the gene level can be used to develop novel control approaches.

### P-24 Teaching old dogs new tricks, or a pilot's perspective on advance flight guidance Del Boyd, bcmvcd@nvwisp.com

Upgrading an aircraft with advanced flight guidance is a challenge for any mosquito district, but adding this upgrade to a 1963 excellently preserved AGCAT<sup>TM</sup> was a particularly challenging and rewarding experience for everyone involved. This poster gives a pilot's perspective of this project with the hope that our experiences will help other mosquito control organizations contemplating similar projects.

### P-25 The species composition and distribution of West Nile virus mosquito vectors in Arlington, VA Aftab Hussain, ahussa@arlingtonva.us, and Muhammad Ahmad

Mosquito surveillance was conducted during the months of May to November from 2004-2007. The goal of this study was to determine the population dynamics of the 10 vector mosquitoes of West Nile virus in Arlington, VA. These species belong to 4 genera: *Culex, Aedes, Anopheles,* and *Ochlerotatus*. The data collected from 2004 to 2007 indicated that *Culex pipiens* and *Cx. restuans* were the most abundant species in Arlington. A decline in *Cx. salinarius* populations was observed in this period. *Aedes albopictus* populations increased consistently during this time, whereas the population of *Ae. aegypti* decreased during this period to almost zero in 2007. The population of other vector mosquito species remained about consistent in this period. Three types of traps (ABC light trap, gravid trap, and BG-sentinel trap) were used to compare their efficacy. The BG-sentinel trap was very effective for catching *Ae. albopictus*.

### P-26 The effects of accumulated organic debris on methoprene efficacy in Ontario catch basins Stacey L. Baker, Stacey.Baker@Ontario.ca, and Norman Yan

The primary pesticide used for control of West Nile virus in Ontario is methoprene, which is applied to over 500,000 catch basins each year. The current application, 2.1% briquet, has not been evaluated in Ontario. A pilot study determined that organic debris in catchments may affect the efficacy of methoprene. In 2005, we tested this possibility with 2 treatments: catch basins (n=17) filled with debris of leaves and sediment and vacuumed catch basins (n=20) free of debris. Catch basins (n=5) with no methoprene treatment were available as controls. As expected, emergence rates approached 100% in the controls. Methoprene was detected more frequently and at higher concentrations in debris-filled catch basins and emergence rates were significantly lower in debris versus cleaned treatments. Vacuumed catch basins had 20.14% (n=5,755) of adults emerge versus 2.69% (n=4,974) in debris-filled catch basins, with difference observed after 26 d. Because methoprene and TOC levels were positively correlated (P<0.001), we attribute the difference in emergence rates primarily to the increased presence of TOC, and to a lesser extent, effects of UV exposure. Methoprene appears to be binding to the organic fraction in water and sediment prolonging higher concentrations, which is reflected in lower mosquito emergence rates.

### P-27 Mortality of non-target arthropods from an aerial application of Evergreen® 60-6 Jonathon Kwan, Jonathan.Kwan@cdph.ca.gov, Mark Novak, Timothy Hyles, and Mike Niemela

Non-target effects from an ultra low volume (ULV) aerial application of pyrethrins were monitored by collecting arthropods from ground tarps placed at the interface of open and canopy areas. A larger number and greater diversity of arthropods were recovered from tarps in the ULV spray area; the observed mortality was approximately 10-fold greater than that in the control area. Arthropods from the treatment area included representatives from 13 orders and >36 families, as compared to 7 orders and >12 families in the control area. Chironomidae (midges) and Formicidae (ants) were the most commonly represented families. Mortality of sentinel mosquitoes in the treatment and control areas averaged 95% and <1%, respectively.

### P-28 Surveillance and control of *Culex* vectors in storm water structures Jon Peterson, jonpeterson@mmcd.org, Kyle Beadle, Nancy Read

Since the introduction of West Nile virus (WNV) to Minnesota, the Metropolitan Mosquito Control District (MMCD) has increased surveillance and control in street catch basins and natural wetlands where vector *Culex* mosquito larvae are found. Storm water management structures built in or near wetlands may have been treated along with other mosquito control operations but have not been identified on our field maps. MMCD's Dakota County field office has worked on a project the past 2 years to map, survey, and provide targeted control in structures classified as culverts, washouts, rip/rap, risers (pond level regulators), and intermittent streams. Field inspectors mapped and inspected over 3,000 structures, and found 2/3 holding water and about 1 in 10 inhabited by mosquito larva at the time they were inspected. Most of the larvae found were of mosquito species known to be enzootic WNV vectors in Minnesota. These types of structures can provide prolific habitat for WNV vectors and their location and treatment is becoming a regular part of the MMCD control program throughout the 7-county metropolitan area.

### P-29 Biochemical characterization of resistant, susceptible, and hybrid *Culex pipiens* (Diptera: Culicidae) strains to *Bacillus sphaericus*

Belal Ahmed Soliman, email address?, Biological and Geological Sciences Dept., Faculty of Education, Suez Canal University, Suez, Egypt, N. M Wassim, and S. M. Khalil

Protein profiles and fractions were detected for *Culex pipiens* (L.) susceptible, resistant strains (F28) to *Bacillus sphaericus* and their offspring resulted from reciprocal crosses (resistant female x susceptible male and vice versa). Proteins were separated on 10% SDS-polyacrylamide gel electrophoresis. Some bands (22 and 32 KD) were common between males, females, and their progeny. Eleven specific bands were restricted to females and 6 bands for males (either susceptible or resistant). Some bands disappeared and a new band (2 KD) appeared in the offspring protein profile. The amino acids sequences deduced from sequence analysis of third domain in 28 S gene of rDNA for susceptible and resistant *Cx. pipiens* strains and their progeny of reciprocal crosses were compared. Three amino acids (arginine, serine, and proline) were repeated with low numbers of repeats in susceptible male and increased in resistant male and vice versa with glycine and leucine amino acids.

### P-30 Aedes aegypti (Linnaeus, 1762) larvae intestinal epithelium changes from Bacillus sphaericus (Neide, 1904) toxic activity

Caroline Dantas de Oliveira, caroline@inpa.gov.br, Fábio Camargo Abdalla, and Wanderli Pedro Tadei

Two *Bacillus sphaericus* strains - S1116 and the standard 2362 - that presented toxic activity to the intestinal track of *Aedes aegypti* larvae, were analyzed. Those strains' toxic effect was assessed at 5, 15, 30, 45 min intervals up to 72 h. The findings revealed that S1116 strain provoked much more intensive cellular responses to the intestinal epithelium, on 5 min to 1 h following contact. When exposed to the 2362 standard strain, the larvae showed total intestinal epithelium cell integrity up to the 72 h reading. In contact with S1116 strain, visible changes on the intestinal epithelium, such as, foregut cell hypertrophy, vacuole presence, release of small vesicles in the intestinal lumen, intracellular separation, and basal membrane cell loosening and rupturing were observed. There was cell disorganization presenting frequent vesicle accumulation both on the ectoperitrophic space and passing through the peritrophic matrix towards the intestinal lumen.

### P-31 Occurrence of Ascogregarina in Manaus/Brasil and its effect in Aedes albopictus and Aedes aegypti larvae in laboratory

Ricardo Augusto dos Passos, ripassos@inpa.gov.br, and Wanderli Pedro Tadei

Occurrence points were registered of Ascogregarina spp. in Aedes albopictus and Ae. aegypti larvae collected in the urban and suburban areas in Manaus/AM. Infected larval percentage and mean infection density were determined. Under laboratory conditions, the weight and survival of Ae. albopictus and Ae. aegypti were similar between pure and mixed cultures; however, the development time of Ae. albopictus was longer in mixed cultures. In pure cultures, only the effect of Ascogregarina taiwanensis on the survival of Ae. albopictus was found. In mixed cultures, a reduction occurred the proportion of Ae. aegypti survivors, in the presence of A. culicis, and in dual infections, 100% Ae. aegypti larva mortality was observed. The weight of both Aedes species was affected by the presence of ascogregarines; nevertheless, the effect of the parasites on the larval development time was not observed. Only the survival of Ae. albopictus was affected by A. taiwanensis in pure cultures. Dual infection provoked high mortality on both species' larvae. A reduction occurred in the weight of both Aedes species in the presence of parasites. The development time of Ae. albopictus and Ae. aegypti was not affected in the presence of Ascogregarina species. Financial support was provided by CAPES/PIATAM.

### P-32 Diflubenzuron insect growth regulator insecticide activity against *Anopheles darlingi* (Diptera: Culicidae) under laboratory conditions

Fábio Medeiros da Costa, fabio@inpa.gov.br, and Wanderli Pedro Tadei

Anopheles darlingi is the main malaria vector in Brasil and is targeted for control. Further research with new insecticides that will enable controlling the vector safely and efficiently is needed. This study aims to assess the larvicidal and pupicidal activity of diflubenzuron, an insect growth regulator, on Anopheles darlingi under laboratory conditions. Diflubenzuron proved to be effective against 4<sup>th</sup> instar larvae and pupae. LC<sub>50</sub> and LC<sub>90</sub> were determined for 4<sup>th</sup> instar larvae at 0.006 and 0.013 ppm, respectively. Pupicidal activity was evident at 0.1 ppm concentration showing that the insecticide also works with 20 min-old pupae being more susceptible than 1-d-old ones. Larvae that were exposed for 24 h presented no significantly different mortality rate than those that had been exposed for 48 h. Diflubenzuron was highly effective against An. darlingi immatures bringing about their mortality and/or morphological alterations leading to the reduction of the emergence of adults.

# P-33 16S rRNA gene sequencing of parasite tick isolated from Anopheles darlingi from Amazon region Thania Virgínia Guaycurus, guaycuru@centroin.com.br, E.N. Assuncão, W.P. Tadei, and S. Astolfi-Filho

This purpose of this preliminary study was to learn more about the tick which parasitizes indoor-resting blood-fed female *Anopheles darlingi*, the major malaria vector in the Amazon region. Each tick was smashed in 50 ml of sterile water. The DNA templates from the suspension were prepared by boiling for 10 min in a Tris-EDTA buffer (20mM Tris, 2mM EDTA, 1% Triton). The 16S rRNA primers used for the PCR experiments were designed in the Laboratory of Recombinant DNA-UFAM. The PCR products for the sequencing were prepared using ExoSAP-IT protocol. The sequencing was performed in capillary sequencer. For the preliminary identification, the 16S rRNA gene sequences were analyzed in BLASTn (http://ncbi.nih.gov/BLAST). Results detected the following microorganisms: Uncultured *Acinetobacter*, *A. calcoaceticus* strain TS2H, *A. rhizosphaerae*, *A. baumannii*, uncultured *Moraxellaceae* bacterium, uncultured bacterium, and glacial ice bacterium. Perhaps this project can open the possibility for the development of experiments using a paratransgenesis-based approach to control malaria transmission in the Amazon region of Brasil.

### P-34 Detection and classification of cry4 mosquitocidal gene of *Bacillus thuringiensis* isolates from Amazon region A. S. Filho, guaycuru@centroin.com.br, E.N. Assunção, S. Astolfi-Filho, and Thania Virgínia Guaycurus

The purpose of this pilot investigation was to search for new strains/genes of *Bacillus thuringiensis* that can be used in mosquito biological control programs in the Amazon region. The PCR approach was performed in 26 B. thuringiensis isolates and involved 2 steps. The first step was the identification of Diptera genes by the use of universal cry4 primer and the second step involved the classification of Diptera specific genes using the cry4 specific primers (cry4A2, cry4B4 and cry cry11A). Of the 26 B. thuringiensis isolates, 10 amplified with the

Diptera cry4 universal primer. By the amplification with the specific cry4 primers, the molecular profiles could be grouped as following: Group 1: 1 strain - cry4A2, cry4B4 and cry11A gene; Group 2: 3 strains - cry11A gene; Group 3: 2 strains - cry4A2 and cry11A gene; Group 4: 1 strain - cry4B4 and cry11A gene; Group 5: 1 strain - cry4A2 gene; and Group 6: 2 strains only cry4 gene. In the preliminary bioassay in *Aedes aegypti* larvae, all the 10 *B. thuringiensis* strains showed high larvicidal activity. PCR results indicated the possibility of the existence another variant of the cry4 gene, which can be verified through the sequencing and phylogeny studies.

### P-35 Identification by PCR of the cry type genes in *B. thuringiensis* isolates from Amazon, Brasil A. S. Filho, guaycuru@centroin.com.br. E. N. Assunção, S. Astolfi-Filho, and Thania Virgínia Guaycurus

The need to characterize novel entomopathogenic proteins led us to search for novel native *Bacillus thuringiensis* from Amazon soil samples. In order to identify the majority of possible of cry genes present in the isolates, we used PCR-based approach, which allows amplification with universal primers (cry1-lep; cry2-lep-dip; cry3; 7; 8-col.and cry4-dip.) of the different cry genes. The DNA was boiled for 10 min and the amplifications were done in a volume of 25 mL, which contained 2,0 mM dNTPs, 2,5 mM MgCl<sub>2</sub>, 1,25U of Taq DNA polymerase, 20 pmol of each primer and 5 ng of DNA. The reaction was cycled for 30 times at 94 °C for 30 s, 50 °C for 30 s and 72 °C for 2 min. The molecular cry type profile of the 26 *B. thuringiensis* isolates were grouped as follows: Group 1: 1 strain – cry 1 gene; Group 2: 4 strains – cry2 gene; Group 3: 10 strains - cry1 and cry2 gene; Group 4: 1 strain – cry2 and cry3 gene; and Group 5: 10 strains- cry 4 gene. The PCR technique in this work enriches current strategies and simplifies the initial stages of large-scale screening of cry genes in *B. thuringiensis* Amazon isolates that contain specific genes or unique combinations of interest with potential insecticidal activities.

### P-36 Mosquitoes (Diptera: Culicidae) associated with West Nile virus transmission in horse stables Luis Ibarra-Juárez, L. Eisen, A. E. Flores, A. Ortega Morales, and I. Fernández Salas, ifernand@fcb.uanl.mx

West Nile virus (WNV) was initially isolated in the New World from mosquitoes and birds in New York City, New York, in 1999. West Nile virus is an arbovirus in the Flaviviridae family which can cause inapparent infection or febrile disease, meningitis, encephalitis and death in birds, humans, and horses. Birds are the main reservoirs and mosquitoes, especially *Culex* species, are the main WNV vectors. Nuevo Leon state is located in northeastern Mexico and is an area where many migratory birds from the US arrive, potentially carrying viruses like WNV. We collected 5,000 mosquitoes using CDC light traps baited with dry ice. Traps were placed in stables in the locality Dulces Nombres, Nuevo Leon, Mexico. The mosquitoes were sorted by species and grouped in pools of 20 mosquitoes each. The species collected were: *Cx. quinquefasciatus, Cx. coronator, Cx. erraticus, Psorophora cyanescens, Ps. ciliata, Coquillettidia perturbans, Aedes vexans, Ae. scapularis, Ae. taeniorhynchus, Ae. albopictus, Anopheles quadrimaculatus, An. pseudopunctipennis, Mansonia dyari, and Uranotaenia lowii. All of the mosquito pools will be processed by Reverse Transcription Polimerase Chain Reaction (RT-PCR) to identify WNV molecular markers.* 

# P-37 Laboratory colonization of Anopheles darlingi and An. benarrochi (Díptera: Culicidae) for experimental infections with parasites of Plasmodium spp. in the Amazon region, Iquitos, Perú Víctor López-Sifuentes, Karín Escobedo-Vargas, Carlos Tong, Johnny Ramírez, David Florin, Joseph Vinetz, victorlopezsifuentes@gmail.com

The inability to colonize *Anopheles darlingi* and *An. benarrochi* is a major limitation that investigators face in developing vaccine strategies in the battle against malaria. In the Peruvian Amazon region, these species represent the main vectors involved in malaria transmission, one of the most serious public health problems in the region. The use of infection-free adult female mosquitoes is a necessary component of many studies pertaining to malaria transmission and vaccine development. In this study, a standardized methodology is presented for the production of female adult mosquitoes (F1) of *An. darlingi* and *An. benarrochi* in the laboratory for the purpose of producing sufficient mosquitoes for experimental infections via the blood of malaria patients. The procedures were developed within the Entomology Program of the Naval Medical Research Center Detachment (NMRCD), located in the city of Iquitos in the northeast region of Peru. The methodology consisted of capturing females in the wild, and then providing stimulation for oviposition. Once the larvae had hatched, an optimum feeding diet was used for the larval development. Finally, a careful process was established for the managing the emerged females who later were used for the experimental infections.

#### P-38 Forensic entomology in Nuevo Leon, Mexico 2006-2007

Humberto Quiroz-Martínez, hqm\_uanl@yahoo.com, Violeta Ariadna Rodríguez-Castro, Daniela Isabel Stephano-Vera, Ricardo Vázquez-Saucedo, Porfirio Díaz Torres, and Eduardo Villagómez-Jasso

Identification of insects associated with human cadavers is relatively uncommon in Mexico. A total of 19 human cadavers with insects present were examined. The insects were collected with forceps and transported in ethanol to the laboratory for curatorial processing. Later, the insects were identified and the information about the conditions in which the cadavers were found was analyzed. The entomologic evidence collected belonged to the orders Diptera, Coleoptera and Hymenoptera, including 9 families, 13 genera, and 10 species, of which the Family Calliphoridae was most diverse in terms of genera and species. Most of the cases studied involved wounds with bleeding, which explains the presence of the great diversity of flies since they are attracted to scents emanated from blood.

#### P-39 Necrophagous insects in the forest ecosystem in Chipinque Ecological Park

Humberto Quiroz-Martínez, hqm\_uanl@yahoo.com, Ariadna Rodriguez Castro, Karla Alejandra Solís Salazar, David Perez Valdez, and Eric Iván Meléndez López

An initial study of necrophagous insects was conducted in a forest habitat in Chipinque Ecological Park using bottle traps with liver as an attractant. Insects were collected weekly; traps were removed and replaced with new ones; and insects collected were separated, counted, and identified in the laboratory. Necrophagous families such as Calliphoridae, Sarcophagidae, Muscidae, Silphidae were identified and included *Lucilia* spp., *Sarcophaga haemorrhoidalis*, *Phannia* spp., *Nicrophagous americanus*, and *Silphus* spp.

#### P-40 Relative repellency efficacy of three formulations against Aedes aegypti

Gabriela Gonzalez, miarana\_@hotmail.com, A. E. Flores, Gustavo Ponce Garcia, and Ildefonso Fernández-Salas

Repellents are an excellent option for short-term reduction of annoyance caused by insects and most importantly to avoid the bites of mosquitoes that transmit vector-borne diseases. It is well known that repellents do not kill mosquitoes; nevertheless, the best products provide long-lasting protection with just 1 application. Laboratory tests were made to compare the efficacy of 3 common commercial formulations, at both low and high concentrations: DEET (7% and 25%), picaridin (5% and 12%), and citronella (5% and 10%). The efficacy of these was tested against laboratory strains of *Aedes aegypti*. Feeding behavior of female mosquitoes was observed following application of each product. Differences were noted between the formulations tested and at different concentrations. An extended-duration repellent formulation containing 25% DEET repelled significantly more than citronella, but not compared with picaridin at 12%. The advantage of picaridin is that it causes less irritation for human skin.

### P-41 Resistance to organophosphate insecticides in *Aedes aegypti* from different municipalities in Aragua state, Venezuela

Enrique Eduardo Pérez Pinto, Henryeduar@gmail.com, and Darjaniva Molina Fernández

The present study determined levels of resistance to organophosphate insecticides in *Aedes aegypti* larvae from 3 municipalities (Girardot, Mario Briceño Iragorri, and Urdaneta) in Aragua State, Venezuela compared with a susceptible strain (Rockefeller) using the WHO immersion method. We evaluated 3 insecticides: malathion, pirimiphos-methyl and temephos. There was resistance to malathion in all strains, with values of FR50 =69.50x for the strain from Girardot, 150.6x for the strain from Mario Briceño Iragorri, and 113.52x for the strain from Urdaneta suggesting this difference in the levels of resistance to this insecticide was caused by the focal nature of its development. The results indicate incipient resistance is developing to pirimifos-methyl, while the temephos is continuously being used in these municipalities and is the best choice for larval control. Evaluating "in vivo" mechanisms of resistance based on oxidase and esterase enzymes, it was found that the former are responsible for resistance to malathion in the Girardot strain, but not in others. This paper suggests studies evaluating resistance at the local level are needed with the aim of achieving better management of insecticides in control programs.

# P-42 Search for mutations in the "super kdr" region of para in *Aedes aegypti* from Latin America G. Reyes-Solís, carmen\_clh@yahoo.com, K. Saavedra-Rodríguez, L. Urdaneta-Márquez, A. E. Flores, I. Fernández-Salas, N. Rodríguez-Neaves, and W.C. Black IV

Pyrethroid resistance in *Aedes aegypti* (Diptera: Culicidae) is increasing worldwide and an insensitive sodium channel is likely to be the major mechanism of resistance. Most of the point mutations conferring knockdown resistance in the insect sodium channel gene occur in hydrophobic segment 6 of domain II of para and are postulated to prevent the rapid paralytic and lethal actions of all known pyrethroids but do not diminish the efficacy of other insecticides. The hydrophobic segment 5 of domain II of para is called "super kdr" and can also carry mutations that confer much greater resistance to DDT and some pyrethroids. We screened exon 19 of para in *Ae. aegypti* that encodes hydrophobic segment 5 of domain II. We examined 1,110 mosquitoes in 37 strains from Latin America and found a transversion in the second position of Leu946 that instead encodes Gln946. We developed a melting curve SNP-PCR assay for these mutations that can be read either on an agarose gel or a melting curve. These results will provide new insight into the mechanisms by which pyrethroids modify the function of voltage sensitive sodium channels.

### P-43 Biochemistry and molecular characterization of the resistance to organosynthetic insecticides by *Anopheles aquasalis* from Rio de Agua, State Sucre, Venezuela

Darjaniva Molina de Fernandez, darja2410@hotmail.com, Luisa Figueroa Acosta, and Marlene Salazar Corredor

The municipality of Libertador, Sucre State in northeastern Venezuela is known as a zone of coastal malaria where the principal vector is *Anopheles aquasalis*. A focal study of insecticide resistance was conducted to determine metabolic resistance mechanisms. Field populations *An. aquasalis* were collected from several localities in Rio de Agua. In the laboratory, we used bioassays and biochemical and molecular tests to identify and document resistance. High levels of alpha and beta esterases, glutathion-s-transferases (GSTs), and inhibited acetylcholinesterase (Ache) activity were detected. The absorbance values for esterases were between 0.8 and 3.5; between 0.19 and 0.32 for the GSTs; and 52.2% for Ache. Mixed function oxidase is not operating because 93.3% of mosquitoes had reference values of less than 0.5 absorbance. Preliminary results with PCR have detected a kdr mutation in the VGSC gene in mosquitoes studied in the locality. Research on *An. aquasalis* resistance is in

progress to improve malaria vector control program in Sucre State and to implement resistance management strategies.

P-44 A historical review of Anopheles (Nyssorhynchus) darlingi distribution in the Peruvian Amazon Roberto Fernández, Roberto.Fernandez@med.navy.mil, Guillermo Calderón F., and David Florin

Anopheles (Nyssorhynchus) darlingi [Root 1926] is one of the major vectors of malaria in the Americas and as such has become a focus of medical entomology research. Raymond C. Shannon is credited with first having documented the species in Peru from specimens collected in 1931 on the Peruvian-Brazilian border. While the original documented collection can be pinpointed, there is a great amount of uncertainty in regards to the past (and present) distribution of this species in Peru. Records indicate the possibility that An. darlingi is a relatively new species inside areas of the Amazon region and that in recent years has greatly expanded its range throughout the Amazon. A thorough review was conducted on all available literature and records to determine past and present distribution within the Peruvian Amazon. Special attention was paid to documenting "nonliterature" sources such as personal communications, internal reports, and conference presentations so that all information up to this date could be accounted for in one publication. Speculation is provided as to the reasons responsible for the apparent dramatic distributional increase of this species in the Peruvian Amazon.

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Zavortink, T.	Bohart Museum of Entomology, University of California, Davis, CA	186
Zazra, D.	North Shore Mosquito Abatement District, Northfield, IL	235
Zhang, A.	USDA, ARS, Invasive Insect Biocontrol and Behavior Laboratory, Beltsville, MD	117
Zhao, L.	CMAVE, ARS-USDA, Gainesville, FL	223, P-23
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#### AWARDS AND OFFICERS

#### AMCA AWARDS

#### HONORARY MEMBERS

1937	Leland O. Howard (USDA)	1965	Arthur W. Lindquist (KS)	1991	Kenneth L. Knight (NC)
1938	C. C. Adams (NY)	1967	Fred L. Stutz (FL)	1994	Harold C. Chapman (LA)
1944	Thomas J. Headlee (NJ)	1970	Robert L. Vannote (NJ)		Lewis T. Nielsen (UT)
	William B. Herms (CA)		Richard W. Fay (USPHS)	1998	Eugene J. Gerberg (MD)
	J. A. LePrince (USPHS)	1971	Christian T. Williamson (NY)		Glen C. Collett (UT)
	Louis L. Williams, Jr. (USPHS)	1972	Alan R. Stone (MD)	1999	Donald R. Johnson (GA)
1948	Robert D. Glascow (NY)		Edward S. Hathaway (LA)	2001	Fred W. Knapp (KY)
	Willard V. King (USDA)	1974	Theodore G. Raley (CA)	2003	E. John Beidler (FL)
1951	Lewis W. Hackett (CA)	1976	John A. Mulrennan, Sr. (FL)	2004	David A. Dame (FL)
	Robert Matheson (NY)	1979	Thomas D. Mulhern (CA)	2005	Donald J. Sutherland (NJ)
1955	Harold F. Gray (CA)		Austin W. Morrill, Jr. (CA)	2006	Martin S. Chomsky (NJ)
1958	Louis A. Stearns (DE)	1981	William R. Horsfall (IL)		
1964	George H. Bradley (USPHS/USDA)	1983	Anthony W. A. Brown (WHO)		

### HAROLD FARNSWORTH GRAY MEMORIAL CITATION MERITORIOUS SERVICE TO MOSQUITO CONTROL AWARD

This discontinued award is to be presented to an active member of AMCA for exceptional service to the Association and to mosquito control or related vector control.

1964

Fred C. Bishopp (DC)

#### DR. THOMAS J. HEADLEE MEMORIAL AWARD

This discontinued award recognizes a living member of the Association for outstanding service to the field of mosquito control, while simultaneously commemorating the name of a deceased member.

1968

George H. Bradley (USDA/USPHS)

#### MEDAL OF HONOR

Next to honorary membership the highest award regularly given by AMCA, the only specific limitation for the Medal of Honor is AMCA membership. Otherwise, nominees are selected on the basis of exceptional contributions to mosquito control or related fields. After 1982, the Board of Directors set a suggested maximum of one Medal of Honor per year.

1972	Maurice W. Provost (FL)	1981	Stanley J. Carpenter (CA)	1993	Gilbert L. Challet (CA)
	William R. Horsfall (IL)		Rowland E. Dorer (VA)	1994	Ronald A. Ward (MD)
1973	Don M. Rees (UT)	1982	Kenneth L. Knight (NC)	1995	T. Wayne Miller (FL)
	Thomas D. Mulhern (CA)		William C. Reeves (CA)	1996	Marshall Laird (New Zealand)
1974	Anthony W. A. Brown (WHO)	1983	Harry D. Pratt (GA)	1997	Robert K. Washino (CA)
	Donald L. Collins (NY)		John A. Mulrennan, Sr. (FL)	1998	John D. Edman (MA)
1975	Daniel M. Jobbins (NJ)	1984	George T. Carmichael (LA)	1999	Bruce F. Eldridge (CA)
	Arthur W. Lindquist (USDA)	1985	Norman G. Gratz (WHO)	2000	Judy A. Hansen (NJ)
1976	Austin W. Morrill, Jr. (CA)	1986	James R. Caton (CA)	2001	Gary G. Clark (USPHS)
	Carroll N. Smith (USDA)	1987	Jay E. Graham (UT)	2002	Lucas G. Terracina (LA)
1978	James B. Kitzmiller (FL)	1988	Lewis T. Nielsen (UT)	2003	Robert J. Novak (IL)
	William D. Murray (CA)	1989	Andrew J. Spielman (MA)	2004	James D. Long (TX)
1979	Richard F. Peters (CA)	1990	Glen C. Collett (UT)	2005	James W. Robinson (FL)
1980	William E. Bickley (MD)	1991	Harold C. Chapman (LA)	2006	John L. Clark Jr. (IL)
	John N. Belkin (CA)	1992	D. Bruce Francy (CO)	2007	E. John Beidler (FL)

Given to individuals for outstanding service, the contributions of the nominees must be considered outstanding as judged by their peers. Only AMCA members in good standing who are not past presidents of AMCA are eligible. After 1982, the Board of Directors set a suggested maximum of no more than 2 awards per year.

1972	Charles F. Scheel (IL)	1976	Donald J. Pletsch (Mexico)	1990	Richard D. Morton (WA)
	Donald L. Collins (NY)		Glenn M. Stokes (LA)		Lucas G. Terracina (LA)
	Theodore G. Raley (CA)		Luis M. Vargas (Mexico)	1991	David A. Dame (FL)
1973	Francis P. Creadon (CA)	1978	Richard C. Axtell (NC)	1992	Jerry Mix (TX)
	Vernon Conant (NJ)	1979	Marco. E. C. Giglioli (BWI)	1993	William E. Hazeltine (CA)
	Austin W. Morrill, Jr. (CA)	1980	James D. Gorman (FL)	1994	Sally A. Wagner (MI)
1974	Leslie D. Beadle (USPHS)		Donald E. Weidhaas (FL)	1995	Frederick W. Wagner (KY)
	John H. Brawley (CA)		E. John Beidler (FL)	1996	Donald J. Sutherland (NJ)
	John W. Kilpatrick (GA)		Eugene J. Gerberg (MD)		Ronald A. Ward (MD)
	T. Oscar Fultz (GA)	1981	A. Ralph Barr (CA)	1997	Roger S. Nasci (CO)
	Howard R. Greenfield (CA)		Gilbert L. Challet (CA)		Thomas J. Zavortink (CA)
	Paul J. Hunt (FL)		Edgar A. Smith (VA)	1998	James D. Long (TX)
	William C. McDuffie (USDA)	1982	Hugo A. Jamnback (NY)	1999	Hilton B. Munns (CA)
	Donald R. Johnson (GA)		Donald R. Johnson (GA)	2000	Leroy J. Bohn (VA)
	Helen Sollers-Riedel (DC)		Harold D. Newsome (MI)		Dreda McCreary (VA)
1975	Lewis E. Fronk (UT)		James V. Smith (GA)	2001	Charles T. Palmisano (LA)
	Joseph G. McWilliams (USN)	1983	Richard F. Darsie (CO)	2002	Thomas G. Floore (FL)
	Lewis J. Ogden (USPHS)		Ronald A. Ward (DC)		Sherry McLaughlin (TX)
	Rajindar M. Pal (WHO)	1984	Samuel G. Breeland (FL)	2003	Wayne L. Kramer (NE)
	Kenneth D. Quarterman (USPHS)		Donald J. Sutherland (NJ)		John L. Clarke, Jr. (IL)
	Herbert F. Schoof (USPHS)	1985	John C. Kuschke (NJ)	2004	Yadira N. Rangel (Venezuela)
1976	Robert A. Armstrong (MA)		James R. Caton (CA)		James W. Robinson (FL)
	Osmond P. Breland (TX)	1986	C. Lamar Meek (LA)	2005	Major S. Dhillon (CA)
	George B. Craig, Jr. (IN)	1987	John C. Combs (CA)		William H. Meredith (DE)
	Claude M. Gjullin (USDA)	1988	Chester G. Moore (CO) Margaret Parsons (OH)	2006	William J. Sames (WA)
	T. Wayne Miller (FL)	1989	John S. Billodeaux (LA) Edgar S. Bordes, Jr. (LA)	2007	Henry R. Rupp (NJ)

#### PRESIDENTIAL CITATION

The Presidential Citation recognizes individuals not eligible to receive other awards but who are eminently deserving of special recognition by AMCA. Recipients need not be AMCA members. After 1982 the Board of Directors set a suggested maximum of no more than 2 awards per year.

1980	John M. Poché (LA) Leslie E. Fronk (UT)	1990 1991	Leonard E. Munsterman (IN) James D. Long (TX)	2000	Peter B. Ghormley (CA) David A. Brown (CA)
	Jesse B. Leslie (NJ)	1992	Charlie D. Morris (FL)	2001	Donald Menard (LA)
1981	Linda G. Raiche (CA)	1993	Robert J. Novak (IL)		Joel Margalit (Israel)
	Margaret S. Slater (NY)	1994	James W. Robinson (FL)	2002	Dennis Moore (FL)
1982	K. G. Nolan (NY)		Dan L. Ariaz (NV)		Henry R. Rupp (NJ)
	Charles F. Scheel (IL)	1995	Sally Kuzenski (LA)	2003	James R. McNelly (NJ)
1983	Coyle E. Knowles (NY)	1996	Carl R. Tanner (IL)		Robert Bonnett (MN)
1984	Ray Treichler (DC)		Sammie L. Dickson (UT)	2004	James R. Brown (FL)
1985	Lawrence T. Cowper (USAID)	1997	Charles T. Palmisano (LA)	2005	Mark Newberg (IL)
	Janice B. Wells (NY)		George J. Wichterman (FL)		Susan Maggy (CA)
1986	T. Oscar Fultz (GA)	1998	Douglas B. Carlson (FL)	2006	Teung Chin
1987	Sharon A. Colvin (IL)	1999	Charles Beesley (CA)	2007	Karl Malamud-Roam
1988	Daniel D. Sprenger (TX)		Donald R. Johnson (GA)		
1989	Fred C. Roberts (CA)				

The John N. Belkin Award is given for meritorious contributions to the field of mosquito systematics and/or biology and may be given to anyone judged by his peers to be worthy. Usually, a maximum of one award per year is given.

1981	Botha de Meillon (PA)	1995	Oswaldo P. Forattini (Brazil)
1982	Lloyd E. Rozeboom (IL)	1996	A. Ralph Barr (CA)
1983	Kenneth L. Knight (NC)		Michael W. Service (UK)
1984	Thomas J. Zavortink (CA)	1997	Christine J. Dahl (Sweden)
1985	Stanley J. Carpenter (CA)	1998	Ralph E. Harbach (UK)
1986	Elizabeth P. Marks & John Reid (Australia)	1999	Yiau-Min Huang (DC)
1987	James B. Kitzmiller (FL)	2000	Lewis T. Nielsen (UT)
1988	Allan R Stone (MD)	2001	John F. Reinert (FL)
1989	Pedro Galindo (Panama)	2002	Richard F. Darsie (FL)
1990	Peter F. Mattingly (UK)	2003	Richard C. Wilkerson (MD)
1991	Jose P. Duret (Argentina)	2004	Kazua Tanaka (Japan)
1992	Bruce A. Harrison (NC)	2005	Ronald A. Ward (MD)
1993	Edward L. Peyton (DC)	2006	William K. Reisen
1994	Theodore H. G. Aitken (CT)		

#### MEMORIAL LECTURE HONOREE & MEMORIAL LECTURER AWARD

The Memorial Lecture Honoree must be one who has made exceptional contributions to the broad field of mosquito control during his lifetime. If there is more than one honoree in a given year, then the group must have made significant contributions as a team or equal stature in the same time frame and to the same aspect of mosquito control. The Memorial Lecturer Award is given to an outstanding speaker (one per year) to present the annual Memorial Lecture in honor of the Memorial Lecture Honoree. The Memorial Lecture Award is not limited to a member of AMCA, but the recipient should be a recognized authority in the broad field of vector control.

	HONOREE	LECTURER	TOPIC
1979	Don M. Rees	J. David Gillett	Out for blood: Flight orientation upwind & in the absence of visual
17/7	Don W. Rees	J. David Gillett	clues
1980	Maurice W. Provost	Anthony W. A. Brown	What have insecticides done for us?
1981	Leland O. Howard	Leonard J. Bruce-Chwatt	Leland Ossian Howard (1857-1950) and malaria control then and now
1982	Carlos Finlay Walter Reed William Gorgas Fred Soper	William C. Reeves	A memorial to Finlay, Reed, Gorgas and Soper as major contributors to present-day concepts essential for control of mosquito-borne viruses
1983	Harry H. Stage	Michael W. Service	Biological control of mosquitoes—Has it a future?
1984	Louis L. Williams	George B. Craig, Jr.	Man-made human disease problems: Tires & LaCrosse virus
1985	Thomas J. Headlee	William R. Horsfall	Mosquito abatement in a changing world
1986	Marston Bates	A. Ralph Barr	The basis of mosquito systematics
1987	William B. Herms	Robert K. Washino	
1988	John A. Mulrennan, Sr.	Susan B. McIver	Mosquitoes, medicine & memories
1989	Brian Hocking	John D. Edman	Are biting flies gourmet or gourmand?
1990	John N. Belkin	Thomas J. Zavortink	Classical taxonomy of mosquitoes—A memorial to John N. Belkin
1991	Edward S. Hathaway Anderson B. Ritter	C. Lamar Meek	Les maringouins du mech: The legacy of two men
1992	Sir Patrick Manson	Bruce F. Eldridge	The man we honor
1993	Willard V. King	Ronald A. Ward	Renaissance man of medical entomology
1994	Stanley B. Freeman	Mir S. Mulla	Now & in the future
1995	Maurice T. James	Wayne A. Rowley	Maurice T. James
1996	Telford H. Work	Charles A. Calisher	Telford H. Work—A tribute
1997	Stanley J. Carpenter	Lewis T. Nielsen	In honor of Stanley Carpenter
1998	George B. Craig, Jr.	Robert J. Novak	George Brownlee Craig
1999	A. Ralph Barr	Andrew J. Spielman	
2000	John B. Smith	Wayne J. Crans	
2001	William R. Horsfall	Jimmy K. Olson	
2002	Edward F. Knipling	Waldemar Klassen	Titan and Driving Force in Ecologically Selective Area-Wide Pest
2003	Kenneth L. Knight	Ralph E. Harbach	Management Mosquito systematics: From organism to molecules—A tribute to Kenneth L. Knight
2004	Donald J. Pletsch	David A. Dame	Six Decades of International Commitment
2005	William E. Hazeltine	Bruce F. Eldridge	William E. Hazeltine: Rebel with a cause

2006	William C. Reeves	Grant R. Campbell	
2007	Norman G. Gratz	Graham B. White	Remembering Norman Gratz (1925-2005) - Doyen of Vector
			Control

#### INDUSTRY AWARD

Established in 1997, the Industry Award is presented to a representative of a mosquito/vector-related industry who has through his efforts advanced the work of mosquito and/or vector control or research.

1997	Charles T. Galley (FL)	2003	Allen W. Wooldridge (FL)
1998	William German (FL)	2004	John L. Clarke, Jr. (IL)
1999	Gary A. Mount (FL)	2005	Ernest Danka (IL)
	Daniel F. Boyd (GA) David W. Waldron (GA)	2006	Willie N. Cox (IL)
	J. David Waldron(GA)	2007	Robert Bonnett (MN)
2002	Robert F. Richard (TX)		

#### DAN F. BOYD AWARD

Established in 2003 in memory of Daniel F. Boyd, this award promotes training for operational mosquito control workers by funding attendance and participation in AMCA-recognized training activities.

#### STUDENT PAPER COMPETITION AWARDS

The AMCA Student Competition was established in 1988 to recognize the outstanding student research paper presented at the annual meeting. Judging of oral presentations is based upon organization, delivery, clarity and effective use of visual aids. In 1991, a \$500 cash award was presented to the winner, and in 1998 the Hollandsworth Prize was established by the family of Gerald Hollandsworth to encourage student participation in the AMCA national meeting. There is a \$250 prize for honorable mention.

1989	Scott Willis	McNeese State U.	2003	Sarah Yaremych	U. Illinois
1990	Andrea Brown	Peru State Coll.		Laura Goddard*	U. California
1991	John Paul Mutebi	Notre Dame U.		Jason L. Rasgon*	U. California, Davis
1992	Rosmarie Kelly	U. Massachusetts	2004	Gregory M. Williams	U. Delaware
1993	Merry L. Holliday-Hanson	U. California, Davis		Stephen Aspen*	Colorado State U.
1994	John E. Gimnig	U. California, Davis		Christian Kaufmann*	U. Zurich
	Alice Shaeffer*	U. Mainz, Germany	2005	Wesley Rubio	San Diego State
1995	Glen Scoles	Notre Dame U.		Whitney Qualls*	Auburn University
	Jittawadee Rochaeroen*	U. California, Riverside		Rebecca Trout*	University of Kentucky
1996	Esther Chow Schaeffer	U. Maryland	2006	Robert D. Anderson	University of Winnipeg
1997	Lynn Cooper	U. Maryland		Linda O'Connor (1st	University of Delaware
1998	C. Roxanne Rutledge	Louisiana State U.		Joshua R. Ogawa*	Oregon State University
	Emmalee Kennedy*	U. Illinois		Matthew Eaton*	Concordia College
	Timothy Schaub*	U. Illinois		Linda M. Styer*	U. California, Davis
1999	Laura Harrington	U. Massachusetts	2007	Jennifer Armistead	University of Florida
	Adam S. Jones*	U. Massachusetts		Robert D. Anderson*	University of Delaware
	Hillary Reno*	U. Illinois		Thomas M. Mascari*	Louisiana State
2000	Jason L. Rasgon	U. California, Davis		Kristen Bartlett*	Rutgers University
	Hope Q. Liu*	Virginia Polytechnic			
2001	No competition	-			
2002	Laura B. Goddard	U. California, Davis			
	Sharon L. Minnick*	U. California, Davis			
	Margaret Sherriffs*	Yale U.			

<sup>\* -</sup> Honorable Mention

#### AMCA OFFICERS, EXECUTIVE DIRECTORS AND EDITORS

#### AMCA PRESIDENTS

1935-1939	Thomas J. Headlee*	1965-1967	Anthony W. A. Brown	1988-1989	Bruce F. Eldridge
1939-1940	Christian T. Williams*	1966-1967	Jay E. Graham	1989-1990	Judy A. Hansen
1940-1942	Louis A. Stearns*	1967-1968	Harry D. Pratt	1990-1991	Robert C. Sjogren
1942-1944	Robert C. Botsford*	1968-1969	Thomas D. Mulhern	1991-1992	Matthew Yates
1944-1945	Robert L. Vannote	1969-1970	George T. Carmichael	1992 -1993	Cyrus R. Lesser
1945-1946	Perry W. Ruth	1970-1971	Albert W. Buzicky	1993-1994	John A. Mulrennan, Jr.
1946-1947	Harry H. Stage	1971-1972	Andrew J. Rogers	1994-1995	Chester G. Moore
1947-1949	H. Duke Peters	1972-1973	Glen C. Collett	1995-1996	John D. Edman
1949-1950	Harold F. Gray	1973-1974	Kenneth L. Knight	1996-1997	Robert J. Novak
1950-1951	Lester W. Smith	1974-1975	Robert M. Altman	1997-1998	Gary G. Clark
1951-1952	Don M. Rees	1975-1976	Harold C. Chapman	1998-1999	Dan L. Ariaz
1952-1953	Cecil R. Twinn	1976-1977	D. Bruce Francy	1999-2000	William J. Zawicki
1953-1954	Fred C. Bishopp	1977-1978	Lewis T. Nielsen	2000-2001	David A. Dame
1955-1956	Rowland F. Dorer	1978-1979	Paul J. Hunt	2001-2002	Sammie L. Dickson
1956-1957	Richard F. Peters	1979-1980	Glen M. Stokes	2002-2003	David A. Brown
1957-1958	Arthur W. Lindquist	1980-1981	Robert K. Washino	2003-2004	Fred W. Knapp
1958-1959	John M. Hirst	1981-1982	Claude H. Schmidt	2004-2005	Roger S. Nasci
1959-1960	Archie D. Hess	1982-1983	Richard C. Axtell	2005-2006	William R. Opp
1960-1961	Daniel M. Jobbins	1983-1984	Jimmy K. Olson	2006-2007	Joseph F. Sanzone
1961-1962	William E. Bickley	1984-1985	Gilbert L. Challet	2007-2008	Gene R. Payne
1962-1963	Arthur W. Geib	1984-1986	T. Oscar Fultz		
1963-1964	Don W. Micks	1986-1987	Donald J. Sutherland		
1964-1965	John A. Mulrennan, Sr.	1987-1988	George B. Craig, Jr.		

<sup>\* -</sup> Eastern Association of Mosquito Control Workers

#### AMCA TREASURERS

1935-1943	Thomas D. Mulhern *
1933-1943	Thomas D. Mumem *
1944-1950	Thomas D. Mulhern
1950-1953	Rowland E. Dorer
1954-1964	Lester W. Smith
1965-1979	William D. Murray
1980-1985	James R. Caton
1985-1986	Douglas C. White
1986-1988	C. Lamar Meek
1989-1994	John S. Billodeaux
1994-2000	Charles T. Palmisano
2000 to date	Allan D. Inman

<sup>\* -</sup> Eastern Association of Mosquito Control Workers

#### SECRETARY, EXECUTIVE SECRETARY, EXECUTIVE DIRECTOR

1935-1943 1944-1950 1950-1952	Thomas D. Mulhern* Thomas D. Mulhern Thomas D. Mulhern	Secretary Secretary Executive Secretary	1986-1991 1991 1992	Harold C. Chapman Lucas G. Terracina Mark Vinsand	Executive Director Acting Executive Executive Director
1953-1973	Theodore G. Raley	Executive Secretary	1992-1993	Harold C. Chapman	Executive Director
1973	Theodore G. Raley	Executive Director	1993-1994	Lucas G. Terracina	Acting Executive
1974-1978	Thomas D. Mulhern	Executive Director	1994-1995	Robert T. Graham	Executive Director
1979-1980	William D. Murray	Executive Director	2006 to date	Sarah B. Gazi	Executive Director
1980-1985	Thomas D. Mulhern	Executive Director			
1985-1986	James R Caton	Interim Executive Director			

<sup>\* -</sup> Eastern Association of Mosquito Control Workers

#### BUSINESS MANAGER

1995-1999	Pamela D. Toups
1999-2000	Marlene Comeaux
2000-2001	Robertamarie Kilev

2001-2004 Martin. S. Chomsky 2004-2006 Sarah B. Gazi

#### TECHNICAL ADVISER

2000 to date Joseph M. Conlon

#### **EDITORS**

#### EDITORS OF MOSQUITO NEWS & JOURNAL OF AMCA\*

1941	Edited by the Publications Committee,
	Lester W. Smith, Chair <sup>†</sup>
1942-1943	Edited by the Publications Committee,
	Ralph W. Vanderwerker, Chair <sup>‡</sup>
1944	Edited by the Publications Committee,
	J. T. Hart, Chair <sup>#</sup>
1944-1948	Robert D. Glascow
1949-1973	Donald L. Collins
1973-1981	William E. Bickley
1981-1996	Ronald A. Ward
1996-1998	Robert K. Washino
1999-2003	Bruce F. Eldridge
2004 to 2006	Kenneth J. Linthicum
2007 - present	Lal S. Mian

#### **EDITORS OF MOSQUITO SYSTEMATICS\***

1969-1979	Kenneth L. Knight
1979-1992	Lewis T. Nielsen
1992-1993	Lewis T. Nielsen &
	Ralph E. Harbach, co-editors
1993-1995 <sup>†</sup>	Thomas J. Zavortink, editor, &
	Lewis T. Nielsen, editor emeritus

<sup>\* -</sup> Prior to 1973 Mosquito Systematics was named Mosquito Systematics Newsletter

<sup>\* -</sup> Mosquito News became the Journal of AMCA in 1985

<sup>† -</sup> Publication of the Eastern Association of Mosquito Control Workers

<sup>&</sup>lt;sup>‡</sup> - Volume 4, Number 1, was edited by the Publications Committee; subsequent volumes had a single editor

 $<sup>^{\</sup>dagger}$  - In 1995 this publication was discontinued

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