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Abstracts

Submitted papers, posters and symposium presentations

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The 2015 AMCA Memorial Lecture Honoree:
Dr. Richard Floyd Darsie, Jr., 1915 - 2014

Dr. Richard Darsie receives an FMCA Presidential Merit Award from Jonathan Day at the 2007 meeting of the Florida Mosquito Control Association.

Richard Floyd Darsie, Jr. was born in Scottdale, Pennsylvania on January 28, 1915. He graduated from Scottdale High School in 1932. In 1937, he was awarded a B.A. degree in biology from Bethany College in West Virginia. He attended the University of Pittsburgh from 1937 to 1941 where he received an M.S. degree in zoology. Dick served in the U.S. Air Force during World War II from 1941 through 1945 after which he resumed his studies at Cornell University where he received a Ph.D. in medical entomology in 1949.

Dick's broad areas of interest and training made him a versatile scientist, teacher, and researcher. The early part of his career was spent at two academic postings, Franklin and Marshall College (1949-1954) and the University of Delaware (1954-1962). During these appointments, Dick was able to perfect a teaching style that would serve him well throughout the remainder of his career. His early research interests focused on mosquito impoundment techniques, new and innovative methods of insecticide application, insecticide resistance, monitoring insecticide treatment efficacy, and non-target impacts of mosquito adulticide and larvicide applications; a suite of interests that would make him an expert in mosquito biology and ecology. More importantly, his interest in adult and immature mosquito morphology and taxonomy, as well as mosquito distribution and bionomics started to become evident during these early years.

From 1962 through 1971, Dick had two international postings, first as a Malaria Specialist stationed at Kathmandu, Nepal for the U.S. Agency for International Development and later as a Training Officer for the Malaria Eradication Training Center in Manila, Philippines. During these appointments, Dick's primary responsibility was to teach adult and immature mosquito identification to students from around the world; an interest and a talent that he would continue to perfect for the rest of his life. It was during this period that the true value of well-designed morphological keys became evident to Dick and he would spend the remainder of his career analyzing and re-analyzing mosquito distribution data and writing and re-writing morphological keys for adult and immature mosquitoes from around the world.

From 1971 through 1976, Dick served as the Chief of Vector-borne Disease Training at the Centers for Disease Control (CDC) in Atlanta, GA. It was during this time that, with Dick's help, the CDC training courses were developed. Courses were offered in the epidemiology and control of vector-borne diseases, mosquito control, and mosquito identification. Dick was the ideal person to teach the mosquito taxonomy and identification sections of these courses.

From 1976 through 1982, Dick moved to Central America where he worked on malaria and onchocerciasis while serving at two international postings. The first was as a Research Entomologist for the CDC Central America Research Station, Bureau of Tropical Diseases in San Salvador, El Salvador. His second posting was as a Research Entomologist for the CDC Medical Entomology Research and Training Unit, Division of Parasitic Diseases in Guatemala City, Guatemala. While in Guatemala, he taught the medical entomology and mosquito identification courses in Spanish, a skill that would be utilized later in his career when he would offer bilingual mosquito identification courses in the United States. From Guatemala City, Dick moved to the CDC laboratory in Ft. Collins, Colorado where he served as a Research Entomologist.

Dick was a Research Entomologist and Adjunct Professor at the International Center for Public Health Research, University of South Carolina in McClellanville from 1985 through 1996 where he taught medical entomology and mosquito identification short courses and conducted research related to the mosquito fauna of Nepal, Guatemala, and Argentina. Dick and his family moved to Vero Beach, FL where he became a visiting Research Scientist from 1996 through 2006 at the University of Florida, Institute of Food and Agricultural Sciences, Florida Medical Entomology Laboratory (FMEL).

Dick Darsie had a remarkable publication record, publishing more than 150 articles and books in peer-reviewed scientific journals. Darsie and Ward (1981, with a Second Edition in 2005), *Identification and Geographical Distribution of the Mosquitoes of North America, North of Mexico* is a publication with which almost every mosquito worker is very familiar. This publication is the major reference for the identification of adult and larval mosquitoes and their distributions in North America.

Dick had a remarkable work ethic and clearly loved all aspects of the study of mosquitoes. In his mid-eighties, he worked five days a week and was often the first to arrive at the laboratory. He was a great collaborator, mentor, and friend to all of us at the FMEL. He seemed to have endless energy and always took a mid-morning walk to exercise his body and clear his head. We all wished we could have bottled some of his energy and enthusiasm and distributed it to the entire laboratory. Dick was tall and thin, yet he had what some would consider a high calorie diet. Every year during Mardi Gras we would order a King cake and Dick would cheerfully partake of at least two large pieces. When mosquito identification courses were offered at the FMEL, students learned on the first day that the first donuts out of the box each morning were for Dr. Darsie. I once asked him how he was able to eat so freely and not gain any weight. He smiled and said, "Good genes."

Few people knew that Dick was an accomplished tenor singer. He sang in church choirs and occasionally at the FMEL. After he learned that there was an equally talented soprano working at the laboratory, the two of them often could be heard singing their favorite hymns including "How Great Thou Art" and "Amazing Grace." Dick was deeply spiritual and as he was preparing to move north in 2006 I asked him about the important life lessons he had learned along the way. His response, "always walk in faith."

When he left Vero Beach, Dick was greatly missed by his friends and colleagues throughout the state, especially by those in professional mosquito control, many of whom learned mosquito identification at the knee of the master. He co-taught the annual FMEL Advanced Mosquito Identification and Certification course with Roxanne Connelly each spring and taught the introduction to adult and larval mosquito identification courses at the Florida Mosquito Control Association's Dodd Short Courses each January.

In Florida, there are many mosquito control professionals who proudly will tell you, "I learned mosquito identification from Dr. Richard Darsie." And that is all that is needed to prove one's credentials in the field, learning the art from the best there is. Roxanne and I were both asked to provide letters of reference for him when he moved to Grove City, PA and sought laboratory space at Grove City College. We found it quite amusing to be asked to provide such a letter for someone with his distinguished resume, but at the same time, we were humbled to write the letters.

His legacy lives on in the hundreds of students across the globe who learned mosquito identification skills from this world-renowned mosquito taxonomist. Even though he is greatly missed, all who knew Dick Darsie should take solace in the joy of celebrating a life well-lived. Richard Floyd Darsie, Jr., one of the greatest mosquito taxonomists of the twentieth century, died peacefully on April 10, 2014 in Grove City, Pennsylvania at the age of 99 after a professional career that spanned eight decades.

The 2015 AMCA Memorial Lecturer:
Jonathan Day

Jonathan Fielding Day was born in Boston, Massachusetts, in 1952. He graduated from Suffield High School in Connecticut in 1970. He completed a Bachelor of Science degree in Environmental Science at Lyndon State College in Lyndonville, Vermont in 1974 where he studied the fleas of small mammals in the boreal forests of northern Vermont and southern Canada. In 1978, he was awarded a Master of Science degree in Ecology at the State University of New York in Fredonia where he studied flea coevolution under Alan H. Benton. He completed a Ph.D. in Medical Entomology at the University of Massachusetts in Amherst in 1981 under John D. Edman and continued his Ph.D. research on the impact of disease infection on host behavior and resulting changes in vector feeding success in the Department of Zoology at the University of Massachusetts. In 1983, he was awarded an NIH R01 grant through the University of Massachusetts and he moved to the University of Florida, Florida Medical Entomology Laboratory (FMEL) in Vero Beach to study mosquito flight, blood feeding behavior, and oviposition in a large outdoor wind tunnel.

In 1986, Jonathan joined the University of Florida faculty as an Assistant Professor stationed at the FMEL where he served as a medical entomologist and ecologist studying the natural history, epidemiology, and ecology of North American mosquito-borne viruses. He was promoted to Associate Professor in 1991 and to Professor in 1998. In 2004, he was named a University of Florida Research Foundation Professor. His main research focus at the FMEL has been on field studies designed to better understand the amplification of arboviruses including St. Louis encephalitis, eastern equine encephalitis, and West Nile viruses. These studies involve tracking the spatial and temporal distribution of arboviruses, mosquito vectors, avian amplification hosts, and the environmental factors that drive these independent biological systems periodically forcing the cycles into synchrony producing conditions where amplification is extremely efficient and results in large numbers of infected mosquitoes and, occasionally, human disease epidemics. His studies also include the use of novel vector and amplification host surveillance techniques, the real time tracking vector populations and avian amplification host populations, predicting epidemic and enzootic transmission, and creating human and domestic animal transmission risk maps that can be viewed at http://eis.ifas.ufl.edu where current and historical model runs are available. In addition, he has conducted research on novel biting fly control strategies including removal trapping protocols directed against saltmarsh mosquitoes and biting midges, mosquito attractants, biting arthropod repellents, and long term avian population changes in south and central Florida.

Jonathan has served as a Subject Editor for the Journal of Medical Entomology since 2002. In 1990, he received a teaching award from the Florida Mosquito Control Association (then the Florida Anti-Mosquito Association) for his teaching and participation in the Dodd Short Courses. In 1992 he was honored with a Research Achievement Award for "outstanding research accomplishments related to the epidemiology of encephalitis" by the Florida Entomological Society. In 2001, he received the United States Department of Agriculture Award for Superior Service, the agency's highest honor, for "outstanding research and education on controlling mosquitoes and other blood-feeding arthropod disease vectors to protect the citizens of Florida and the Nation." In 2004, he was awarded a Science and Engineering Visualization Challenge Award from the National Science Foundation and the Journal Science. The Visualization Challenge Award is "an international completion to recognize outstanding achievements by scientist, engineers, visualization specialist and artists who are innovators in using visual media to promote understanding of research results and scientific phenomena." In 2005, he was awarded the Joseph Y. Porter Distinguished Achievement Award by the Florida Mosquito Control Association in recognition of his contributions to entomology with special emphasis on the abatement of arthropods of public health importance. In 2006, he served as the President of the Society for Vector Ecology. Jonathan and his coauthors have published more than 115 peer-reviewed publications including ten book chapters.

Jonathan lives with his wife Marianne in Vero Beach, Florida. They have four children: Ashley, Alyse, Michaelene, and Spencer and four grandchildren: Jillian, Mia, Beckham, and Brooks with a fifth on the way. Much of Jonathan's free time is spent volunteering. He teaches biology and a senior thesis seminar at a local high school, has been appointed to serve on the Indian River County Planning and Zoning Commission since 2011, and is a Eucharistic Minister in the Anglican Church.

Oral Presentation Abstracts

Plenary Session

PL-1 Yes I Can! Making a Difference through a Positive Mindset
Rick Labell

Yes I Can! Three little words made up of seven little letters that can make all the difference in the world! A positive attitude will drive us to make the right choices in life, it will determine whether we succeed or we fail, it's the governing factor on how we approach our daily lives, both personally and professionally and it's that positive way of thinking that will propel us into making a difference in the lives of other people and the society in which we live. I know firsthand, because it is those three little words that allowed my family to survive the devastating loss of our daughter Kelly from the cureless disease of eastern equine encephalitis. This presentation will improve your life, it will enhance your work ethic and most importantly it will inspire you to continue what you do in order to make this world a better place for all of us to live. You'll walk away empowered, refreshed and ready to conquer!

PL-2 Challenges in Public Health Entomology
Jerome Goddard, Ph.D, jgoddard@entomology.msstate.edu

Historically, the discipline of public health entomology was concerned primarily with filth fly and mosquito control, with special emphasis on the diseases transmitted by these vectors (e.g., malaria). Twenty-first century public health entomology has been pre-occupied with Lyme disease, West Nile virus, dengue, chikungunya, and of course, malaria. Challenges facing persons charged with public health entomology responsibilities include lack of infrastructure in developing countries, anti-pesticide fears in first-world, developed countries, immigration (both legal and illegal), movement of invasive vector species, and financial conflicts of interests tainting scientific research and product development. This presentation will address each of these challenges and discuss strategies for dealing with them.

Chikungunya: An Emerging Threat to the Americas I

1 Introduction to the symposium
Roger Nasci, RNasci@cdc.gov

Chikungunya virus (CHIKV) entered the Western Hemisphere in late 2013 and within a few months caused over 100,000 cases extending over 15 Caribbean island countries and 2 South American countries. As a result, the number of chikungunya-infected travelers returning to the United States increased, expanding the risk of local transmission on the US mainland. This symposium will describe the global chikungunya epidemic, epidemiology of chikungunya as it spread in the Western Hemisphere, the experience of 1 state health department in conducting chikungunya surveillance, and observations about controlling *Aedes aegypti* and *Ae. albopictus* from 2 mosquito control programs in areas at high risk for CHIKV introduction and establishment.

2 Chikungunya - the global epidemic
Ann Powers, APowers@cdc.gov

Chikungunya virus (CHIKV) is a mosquito-borne virus that has been responsible for over 3 million human infections during the past decade. Prior to 2006, CHIKV was a little known agent outside its enzootic distribution and the few sites of epidemics. This virus, which previously had a geographic range primarily restricted to Sub-Saharan Africa, the Indian subcontinent, and Southeast Asia, has greatly expanded its range in a series of ongoing outbreaks during which time the virus has moved to subtropical latitudes as well as novel ecosystems. This expansion was not completely unexpected given the characteristics of the virus and the nature of the epidemics. This presentation will describe the movement of the virus over the past 10 years and describe the risks that made further expansion inevitable.

3 Chikungunya in the Western Hemisphere
Roger Nasci, RNasci@cdc.gov

The number of chikungunya-infected travelers entering the Americas, including the United States, increased in response to the global expansion of the virus that began in 2004. In anticipation of the inevitable introduction of the virus to the Western Hemisphere, regional health agencies (CDC, PAHO, CARPHA and others) initiated efforts to prepare the region to detect and respond to chikungunya. In December 2013, the first locally-transmitted cases were reported from St. Martin. In the following months, infected travelers distributed the virus rapidly through the region. Within 9 mo of the first reported cases, over 500,000 locally-acquired cases were reported from 33 countries (25 in the Caribbean islands, 3 in Central America, 4

in northern South America, and 1 in North America – Florida, USA). This presentation will describe US and regional preparations for chikungunya, the increase in traveler-introductions, the rapid course of the regional outbreak, and the risk chikungunya poses to the United States.

4 Chikungunya fever surveillance, Florida, 2014

Carina Blackmore, Carina.Blackmore@FLHealth.gov, Katherine Kendrick, Lea Heberlein-Larson and Danielle Stanek

On June 27, 2014, the Florida Department of Health in Miami-Dade County was notified of a chikungunya fever patient with no recent travel to an endemic area. After careful investigation and additional testing, this patient was confirmed as the first locally-acquired chikungunya fever case in the continental United States.

Soon after the first local transmission of chikungunya virus was reported in the Americas in December 2013, the Florida Department of Health enhanced chikungunya fever surveillance activities. Efforts included outreach to health care providers about the clinical presentation of the disease and the availability of PCR, and antibody testing at our State Public Health Laboratory. As of October 25, 2014, 313 travel-associated chikungunya fever cases have been reported. Given the large number of travelers infected with chikungunya fever and the wide spread distribution of *Aedes aegypti* and *Ae. albopictus*, local transmission of the virus in Florida is not surprising.

To date, Florida has also reported 11 locally-acquired cases from 4 South Florida counties. Three of these (Miami-Dade, Palm Beach, and Broward counties) have reported 146 (47%) of the 313 imported cases in Florida. Two cases appear to be linked; the source is unknown. Among imported cases, the most common country of exposure was Haiti (33%); the most common reason for travel was to visit friends and relatives.

Surveillance related activities included enhanced syndromic surveillance, medical record review, media coverage, reverse 911 dialing, and targeted mailings. For over half of the cases, both local and imported, local mosquito control deployed to cases' residences the same day as or before counties received positive laboratory test results.

5 Controlling *Aedes aegypti*

Michael S. Doyle, mdoyle@keysmosquito.org

Controlling *Aedes aegypti* requires approaches not common in the US. Control presuppositions generally revolve around floodwater nuisance mosquitoes, focusing on geographic efficiency. Most current *Ae. aegypti* strategies require nearly the opposite approach.

Aedes aegypti adults are primarily day-time biters, harboring either in highly sheltered areas near humans. Individual females often bite numerous humans to obtain a bloodmeal, allowing for low densities to create nuisance problems, and to transmit disease. Their affinity for humans has exposed multiple generations to common insecticides, resulting in high rates of resistance in many areas.

Aedes aegypti eggs and larvae are hidden in thousands of small containers in, around, and under houses. The larvae do not require heavy organic matter, making nearly any solid material around the home a potential larval site. Common first approaches attempt to treat massive numbers of containers one-by-one, or attempt to adapt large-scale larviciding methods.

These approaches can cost a great deal in manpower, overhead, insecticide costs, and shifts in employee expectations. Treating containers requires many inspectors per square mile, requires large investments in staff, trucks, and equipment. In addition, the sheer number of larval sites (i.e., containers or homes) requires high data management overhead, hours of public interaction, and often private property complications.

Options for future control methods will be compared and discussed: container removal/dumping, specialized truck and aerial larviciding, barrier treatments (including ATSB), thermal fogging, and self-disseminating strategies such as pyriproxyfen, *Wolbachia* infection, and genetic modification.

Genetics/GIS

6 Multiplex qPCR assay detects genomic copy numbers of two esterase genes associated with insecticide resistance in *Culex pipiens* complex mosquitoes in the US

Linda Kothera, fph6@cdc.gov, Enas Ghallab, Janet McAllister and Harry Savage

Insecticide resistance in vector mosquitoes can be the result of a variety of changes to genes and metabolic functions. In *Culex pipiens* complex mosquitoes, 2 esterase genes (CPIJ013917 and CPIJ013918) confer resistance to organophosphate (OP) insecticides either through upregulation or gene amplification. Previous work to quantify gene copy numbers has examined the 2 genes separately, or considered only 1 of the

members of the *Cx. pipiens* complex. We sought to develop an assay that examines copy numbers of both target genes simultaneously and works in both *Cx. pipiens* and *Cx. quinquefasciatus* in the US. The resulting real-time PCR assay uses primer pairs with fluorescent probes to calculate genomic copies of the target genes relative to the known single-copy reference gene Ace2 (CPIJ000662). Starting with DNA from pools of 10 Sebring colony individuals, we created standard curves for each of the 3 genes separately, and then as a multiplex. After verifying that the levels of efficiency and reproducibility were acceptable, each of the 3 amplicons was cloned using TOPO-TA cloning kits to create single-copy plasmid DNA, which was mixed and run on every plate in triplicate as a positive control. The assay was tested on DNA from adults raised from field-collected egg rafts whose resistance levels to malathion and permethrin had been assessed with a bottle bioassay. The number of gene copies was variable within the populations tested. In addition, there was a positive correlation between copy numbers of each target gene and levels of resistance.

7 Evolutionary genetics of the partial immune genes (TOLL6 and CLIPA6) in *Anopheles minimus* mosquito

Uraiwan Arunyawat, uraiwan.a@ku.ac.th, Prin Phunngam and Theeraphap Chareonviriyaphap

Understanding evolutionary processes that govern susceptibility in mosquito vectors would have important public health implications for vector-borne diseases such as malaria. If the mosquito innate immune system is used as a principle blocking step in malaria transmission, then the immunity genes might have evolved under selection forces imposed by malaria parasite transmission. In this study, 2 partial immune genes including *TOLL6,* which involves signal transduction pathways of insect immune system, and *CLIPA6* involving signal modulation and amplification pathways were selected to estimate patterns of genetic diversity in *Anopheles minimus*, the main malaria vector in Southeast Asia. Primers were designed and tested to amplify the *immune gene* fragments, and the PCR products were directly sequenced and analyzed using single nucleotide polymorphism data. Low levels of nucleotide diversity were shown in the studied *An. minimus* populations. Statistical neutrality tests, including Tajima's *D* and Fu and Li's *D**, were used to measure deviation from neutral equilibrium expectation. No significant negative Tajima's *D* and Fu and Li's *D** values were detected in this study, indicating an excess of low frequency polymorphisms. The significance of mismatch distribution tests supported the population expansion model. In addition, Ka/Ks ration test indicated that the variation of the partial *CLIPA6* and *TOLL6* is most likely explained by the influence of mutation and genetic-drift interaction rather than the effect of directional selection.

8 Pyrethroid resistance and *kdr* mutations are widespread among Florida populations of *Aedes aegypti*

Alden Estep, alden.estep@ars.usda.gov, Neil Sanscrainte and James Becnel

Aedes aegypti is an efficient vector of a number of diseases that affect man and is of increasing concern because of the reemergence of dengue and recent identification of locally acquired chikungunya in Florida. Pesticide resistance in this species has been demonstrated in several neighboring countries throughout the Caribbean. The resistance status of local populations is of critical importance for effective management by control districts in Florida but little is known. In this study, we performed toxicological testing by direct topical application on populations of *Ae. aegypti* collected from several locations in north and central Florida. These populations showed up to 85-fold resistance to permethrin when compared to the susceptible Orlando-1952 strain. Resistance levels in some wildtype populations were similar to those of a Puerto Rican pyrethroid-resistant line available from BEI Resources. We examined the molecular basis for this resistance by performing melt curve analysis of the sodium channel transcript to determine the presence of the *kdr* mutation. This mutation is widespread in the populations we examined but at various levels of penetrance. Assays with synergist examined the contribution of esterase activity to resistance in each population.

9 Intron variation in *Timeless* gene from *Anopheles* mosquitoes in Thailand

Thananya Comproh, s_sungsit@hotmail.com, Sungsit Sungvornyothin, Uraiwan Arunyawat, Theeraphap Chareonviriyaphap and Waraporn Juntarajumnong

A variety of critical behaviors of *Anopheles* mosquitoes related to malaria transmission such as flight activity, host seeking, blood feeding and egg laying have been controlled by biochemical pathways of circadian genes. Among the pathway complexes, *Timeless* gene has been found to be the center of circadian mechanism which connects the endogenous clock with the external environment; for example, changing of temperature and light intensity due to climate change and global warming situation. Unfortunately, the recent information of *Timeless* gene of *Anopheles* mosquitoes in Asia is very scarce. The representatives of 4 groups of major vectors in Thailand (*dirus, minimus, maculatus* and *sundaicus*) were collected from cattle baited method and were preserved in ethanol to study molecular basis on short sequence of *Timeless* gene. PCR primers for this region were designed from the *Timeless* mRNA sequence database of some dipteran species. Successful polymerase chain reaction of *Timeless* gene was sequenced and bioinformatic analyzed. DNA sequences were high consensus to mRNA sequence from *Anopheles gambiae* except an interval of 75 – 78 bp which were interpreted to be an intron region. The intron sequences belonging to the *An. dirus* group contained 9 - 11 repeats of CAG microsatellites. Phylogenetic analysis of PCR product sequences showed

that variation of intron sequence and length was the major factor of clade grouping. This is the first investigation on *Timeless* DNA structure and microsatellite- intron discovery from the important malaria vectors of Asia.

10 Building on the Esri ArcGIS technology platform

Ryan Pierson, ryan@elecdata.com and Chad Minteer

As the use of GIS technology expands throughout the vector control community, it is important to understand all of the benefits and functionality of any GIS platform. The Esri ArcGIS technology platform includes much more capability than merely the commonly used desktop ArcMap GIS software application. Whether the needs of the organization include software development, mobile application support, monitoring operational status, or engaging the public, the Esri ArcGIS platform provides specific functionality to address the needs of any organization. This presentation will demonstrate the use of the Esri ArcGIS technology platform to support the office staff, empower the field workers, engage the public, and inform the program managers in a typical vector control organization. Specifically this presentation will demonstrate the Esri ArcGIS platform tools for easily and quickly creating custom web and mobile mapping applications, publishing GIS content, delivering published GIS content to the entire organization and to the public, creating executive dashboards to monitor daily activities, deploying applications to mobile devices to support field staff, and managing operational data and maps.

Although our software products, Sentinel GIS and FieldSeeker GIS, are built upon this Esri ArcGIS technology platform, this presentation will focus more upon the functionality, tools, and capability that make the ArcGIS technology platform a very valuable asset to any vector control organization. This includes demonstrating portions of our vector control software on Apple iOS devices as used by field technicians.

11 Utilization of GIS in surveillance and control of disease vector mosquitoes in Harris County, Texas

Vence Salvato, vsalvato@hcphes.org, Maximea Vigilant, Yvonne Randle, Lauren Wilkerson, Martin Reyna, Kyle Flatt and Mustapha Debboun

The introduction of West Nile virus in 2002 resulted in the expansion of Harris County Public Health and Environmental Services (HCPHES) - Mosquito Control Division's utilization of GIS technologies. This gave HCPHES-Mosquito Control the ability to plot and analyze their surveillance (mosquito and bird) and treatment (ground ULV and aerial) data, which allows them to visually interpret their data and aids in decision-making processes. All program elements have benefited from these technologies that have streamlined data collection, storage, analysis, and sharing between internal and external stakeholders. This presentation focuses on the record breaking 2014 West Nile virus season and the use of GIS in surveillance and control throughout the season.

12 DropVision for adulticide and larvicide measurements

Bill Reynolds, breynolds@leateam.com

DropVision has been integrating into many mosquito control operations throughout the United States and Mexico, both in the private and public sector. Learn the importance and significance of knowing the droplet spectrums being produced by ground or aerial application equipment used in your operation. Gain knowledge of the many useful reasons for making DropVision a part of your organization.

13 Leading Edge Satloc G4 Integrated GPS/GIS System

Bill Reynolds, breynolds@leateam.com

The Satloc G4 is the newest GPS guidance and recording system available and has been used operationally in aerial vector control for 2 yr. More than 2,000 systems have been deployed world wide in the agriculture and forestry markets. Leading Edge has designed and integrated a web-based GIS solution within MapVision and as a stand alone system, providing the user with easy operation of the G4 features. The G4 also performs drift optimization in real time for aerial adulticiding and larviciding applications.

14 MapVision enhancements

Joel Buettner, joelb@placermosquito.org

Introduced in 2008, MapVision has evolved into a comprehensive, powerful enterprise system. Leading Edge continually reinvests into innovative enhancements. This presentation will highlight new product features and the user benefits.

New Product Trials

15 Effective, efficient, economical, & environmentally friendly: Consistently proven test results in Brazil using Bite Back! Mosquito larva traps

Julio Abreu, julio_abreu13@hotmail.com, James Forehand and Ricardo Guerra
Bite Back! is an innovative mosquito control device designed to trap mosquito larvae, thus breaking the exponential growth cycle. Data were collected on the 3rd phase of a 12-mo test in a mapped, residential area of 436 single family homes and a neighboring commercial area of approximately 319,900 m². Results obtained when Bite Back was deployed in conjunction with other control methods like larvicide treatments and residual and ULV space spray applications were outstanding. Tests results were exceedingly consistent and will pave the way for BiteBack's inclusion in mosquito control operations around the world.

The Bite Back trap is cost effective, simple and efficient, and it can also easily fit into the sustainability category, as it is uncomplicated to manage, environmentally safe, and uses no poison.

The Bite Back concept was developed and patented by Mr. Jimmy Forehand from Atlanta, GA, and officially tested by the University of Florida. I brought a sample back to Brazil to be tested by my company, and the results were outstanding. Certain that we were on the forefront of a great innovation, in 2010 we imported 6,000 kits, or 24,000 traps for use in further studies and distribution. This led to tests in Brazil by the local Brasília Health Public Agency, again with an exceedingly positive outcome.

To obtain optimum mosquito control, Bite Back should be applied in combination with the other control methods involved in the actions already known in the existing mosquito integrated control as per WHO Lab and Field Guidelines.

16 Comparison of ground-applied ULV deltamethrin (DeltaGard Public Health Insecticide) to ULV etofenprox (Zenivex® E20) on *Culex quinquefasciatus* and *Cx. tarsalis*

Kirk Smith, ksmith@mail.maricopa.gov and Hans Olsen

Bayer Environmental Science will be introducing a new ULV mosquito adulticide product in 2015. In September 2014, a field study was conducted near Phoenix, AZ in cooperation with the Maricopa County Environmental Services Department Vector Control Division. This study was designed to compare the efficacy of DeltaGard Public Health Insecticide with Zenivex® E20 using a single-pass caged mosquito ULV ground-fogger trial. DeltaGard PHI was applied at 0.00089 lb AI/acre and Zenivex® E20 at 0.00350 lb AI/acre in separate truck mounted Curtis Dyna-Jet L30 electric foggers. The products were applied from an 800-ft driveline positioned upwind 100 ft from a 3 x 3 grid of stations with 100-ft spacing. There were 25 to 30 mixed wild caught mosquitoes per cage with 2 cages per station. Mortality at 24 h for DeltaGard PHI was 99.8% and Zenivex® E20 was 55.8%. A second Zenivex® E20 trial was conducted 2 wk later and mortality at 24 h was 90.9%. Potential reasons for the Zenivex discrepancies will be discussed.

17 2014 summer trial using FourStar *Bti*-CRG in irrigated pastures

David Heft, dheft@fire2wire.com
During the summer of 2014, Turlock Mosquito Abatement District conducted a field trial of FourStar's *Bti*-CRG product on flood irrigated pastures. FourStar *Bti*-CRG is a heavy, high-density and sand-based larvicide designed originally for saltwater habitats with vegetative canopy. Dual action controlled release technology allows FourStar *Bti*-CRG to remain effective through wet and dry periods (i.e., tidal flows). This trial was aimed at testing whether this same technology could be effective in controlling mosquitoes in flood-irrigated pastures which are flooded with water every 14-20 days. The ability of this product to withstand periodic wet and dry periods coupled with its active ingredient (*Bacillus thuringiensis)* means this product could prove to be a very valuable rotational chemical in treating irrigated pastures. *Bti*-CRG was applied to several pasture locations and post-application measured both larval and adult activity. Subsequent irrigation dates were recorded and inspections continued to measure how many days and/or irrigation cycles the product remained effective. Inspection data reveal that the product remains highly effective under normal conditions through 3 irrigation cycles and was reasonably effective through 4. During the hot summer months, effectiveness of the product was decreased at the 10 lb/acre rate due perhaps to the slow release nature of the product and the fast development rate of mosquitoes such as *Aedes nigromaculis*. Repeated trials at the 20 lb/acre rate during the hot summer months proved to be effective for 3-4 irrigation cycles (40+ d).

18 Efficacy of a commercial formulation of attractive toxic sugar baits (ATSB) against *Aedes albopictus*

Amy Junnila, amyjunnila@gmail.com, Vasiliy Kravchenko, Edita Revay, Yosef Schlein, Whitney Qualls, Rui-de Xue, John Beier and Gunter Muller

We tested the efficacy of an attractive toxic sugar bait (ATSB) solution, with garlic oil microencapsulated in beta-cyclodextrin as the active ingredient, against *Aedes albopictus* in suburban Haifa, Israel. Two, 3-acre gardens with high numbers of *Ae. albopictus* were chosen for perimeter spray treatment. The experimental site received ATSB while the control site received attractive sugar bait (ASB) which is the same as ATSB but without toxin. Baits were colored with food dye to verify feeding of the mosquitoes. The population was monitored by human landing catches and sweep net catches in the surrounding vegetation. Treatment occurred on day 13 of a 44-day monitoring period.

The mosquito population collapsed about 4 days after treatment and continued to drop steadily for 27 days until the end of the study. Pre-treatment, at the experimental site, the average biting pressure was 17.2 per 5 min. Two days post-treatment, the biting pressure dropped to 11.44, and continued to drop to an average of 2.58 during the following 26 days. At the same time the control population was stable. Few sugar fed females (8 to 10%) approached a human bait and anthrone tests showed relatively small amounts of sugar within their crop/gut. Around 60 to 70% of males caught near human bait were sugar positive which may indicate that males were feeding on sugar for mating related behavior. From the vegetation treated with the toxic bait, we recovered significantly less (about 10 to 14%) mosquitoes stained by ATSB than ASB. This could indicate that the toxic baits may also change the resting behavior of poisoned mosquitoes within the vegetation. Almost no *Ae. albopictus* females (5.22 ± 1.38) approached human bait after they fed upon ATSB.

19 Nationwide field efficacy trials of DeltaGard Public Health Insecticide

Kurt Vandock, kurt.vandock@bayer.com and Gordon Morrison

DeltaGard Public Health Insecticide represents the introduction of a new active ingredient (deltamethrin) for wide area mosquito control in the United States. Throughout 2014, field trials were conducted in 18 states, representing more than 20 cooperating mosquito control districts. Over 26 different species of mosquitoes were utilized for these trials, many of which with documented resistance. Beyond characterization of DeltaGard Public Health Insecticide, 8 major adulticides with various active ingredients were assessed for comparative efficacy. We will present results from these trials, highlighting comparative efficacy, performance against resistant strains, and an assessment of application rates.

20 Swimming pool efficacy trials with CocoBear™ Mosquito Larvicide Oil and Natular™ Tablets in Maricopa County, Arizona

Kirk Smith, ksmith@mail.maricopa.gov

Residential swimming pools continue to be a major component of our mosquito management program. The greater Phoenix area still ranks in the top 2 for foreclosed and/or abandoned properties. From 2013 to the present, we have inspected approximately 7,000 pools for mosquitoes, and served over 400 warrants for access to properties when a responsible party could not be discovered. *Culex quinquefasciatus* is the predominant species. A variety of larvicide products are used for our abatement program. For this report, we will present data and observations regarding our testing of two different products, CocoBear™ Oil and Natular™ Tablets

21 Field evaluation of DeltaGard (deltamethrin) as a new ground-based ULV alternative

Daniel Markowski, dmarkowski@vdci.net and Kris New

We compared the performance of DeltaGard applied at low, mid, and high label rates to 7 competitive products using populations of adult mosquitoes (*Anopheles quadrimaculatus*) in Greenville, MS, by assessing mortality with standard caged trials. The trials yielded very favorable results for DeltaGard (deltamethrin), especially at the higher label rates. We observed an overall mortality of 98.3% and 98.9% at the mid and high label rates, respectively. We also observed very good and consistent mortality at all cage distances. Our best performing product in this region of the country has historically been Scourge (resmethrin). And, again, Scourge 4-12 performed very well with an overall mortality of 96.4% applied at 0.007 lb AI/acre. Permanone RTU (permethrin) produced an overall mortality of 91.0% at the same label rate. The results for the Anvil (sumithrin), Duet (sumithrin, prallethrin) and Zenivex (etofenprox) trials at their highest label rates provided expected results for our area with an overall mortality of 85.8, 76.5 and 75.9%, respectively. At its mid-label rate (0.0008 lb AI/acre) DeltaGard performed as well as Scourge at a maximum label rate (0.007lb AI/acre). In conclusion, DeltaGard performed very well against a very difficult to control mosquito with known tolerance to several mosquito control adulticides.

22 Field trials involved in the registration of an organic-approved adulticide Merus™ 2.0
Derek Drews, ddrews@clarke.com, Jacob Hartle and Grifith Lizarraga

From 1994 to 2013 the number of organic farms in the United States has increased from 2,500 to over 18,000. This 7-fold increase in the number of organic farms has restricted for some mosquito abatement programs the use of conventional adulticides in and around these lands. An Organic Materials Review Institute (OMRI) listed adulticide is essential for an integrated mosquito management plan to control disease and nuisance mosquitoes in these areas. Merus™ 2.0, an OMRI-listed adulticide containing 5% pyrethrin, has been approved for use in and around organic farms, giving the industry a tool to use in environmentally sensitive areas. From 2012-2013 Merus™ 2.0 moved from lab through field trial assessments against adults of 3 mosquito species. At a 0.5 fluid oz/acre application rate, with a volume mean diameter range of 10.5 to 15.4 microns, Merus 2.0 killed an average of 99% *Aedes aegypti* (622/623), 96% *Anopheles quadrimaculatus* (554/579), and 100 % *Culex quinquefasciatus* (643/643) 24 h after treatment at a field site in Lake Wales, FL.

23 Functional micro-dispensers (FMDs) as the next generation control against mosquitoes
Noel M. Elman, nelman@mit.edu, Hilla Conen, Oscar R. Miranda, Ulrich R. Bernier, Gary Clark, Daniel L. Kline, Joyce Urban and Sebastian D'hers

Functional micro-dispensers (FMDs) were developed for controlled release of insecticides for optimized spatial coverage, maximized protection and limited toxicity to humans. Computational Fluid Dynamics (CFD) models were utilized for optimizing design taking into account physical properties of the device and chemicals, e.g., micropores, surface area of evaporation, evaporation rate and wettability of materials. FMDs were fabricated using state-of-the-art SLA 3D printing, defining polymeric structures with micrometer resolution. Two sets of experiments were performed to test efficacy against *Aedes aegypti*. The first experiment consisted of testing knock-down of mosquitoes. Experimental results show that the FMDs were effective to knock-down mosquitoes using only metofluthrin, obtaining 99% mortality within 12 h. Additional experimental results showed that a combination of metofluthrin, transfluthrin, 1-Methylpiperazine, DEET, obtained a mortality of 98% within 12 h. The second set of experiments was performed to test repellency. This test consisted of using a mosquito magnet based on a continuous source of CO_2 to attract mosquitoes inside of the open tent. The tent was located inside of a large caged structure in which approximately 1,000 mosquitoes were released and confined to fly. The control was defined as 30% attraction, as approximately only 300 mosquitoes entered the tent attracted by the magnet. Experimental results showed that the FMDs exhibited approximately 70% repellency for 1-Methylpiperazine and Niaouli relative to the control. FMDs promise to achieve large social impact for improved and cost-effective protection in military and civilian settings.

Regulatory/Non Target/Education

24 A global perspective of regulatory and safety requirements for registration of a non DEET insect repellent ethyl butlacetylaminopropionate (IR3535)
Howard Epstein, howard.epstein@emdmillipore.com and John DeBlasi

Insect repellents are regulated because they repel or mitigate pests. By definition they meet the criteria for a pesticide/biocide. These products are generally applied to skin and users may be exposed to the AI for prolonged periods of time or by repeated application over hours, days and even weeks. In 1973, the World Health Organization started to develop a tentative classification of pesticides that would distinguish between the more and less hazardous forms of each pesticide. Industry must ensure each pesticide product is adequately and effectively tested by recognized procedures and test methods for safety and efficacy. Governments need to establish pesticide registration schemes that ensure all relevant safety and efficacy data are available to their citizens to minimize risk in use and ensure the product is effective. This presentation will review global differences and similarities experienced when registering IR3535 as a pesticide in different regions of the world. Greater international harmonization of pesticide data requirements reduces the need for duplicative testing by industry and saves resources while reducing time to register, less cost and improved efficacy of product performance.

25 Aligning mosquito control operations and pollinator protection
Stephen Manweiler, mmcd-sam@mmcd.org and Michael McLean

Mosquito control operations are not incompatible with protecting pollinators and other non-target organisms. Understanding issues of citizens to whom services are provided and communicating how operations include measures to preserve pollinators is critical. Mosquito control operations can be viewed as preserving the environment while protecting public health.

26 The Fourth Amendment and the Orange County Vector Control District's efforts at controlling a record year of West Nile virus activity via enhanced inspection and treatment of larval mosquito sources on private property

Colin Burns, crburns@harperburns.com, Robert Cummings, Steve Koenig, John Newton and Larry Shaw

Controlling West Nile and other mosquito-borne viruses requires aggressive inspection and treatment. Continual, repetitive inspections of private property to identify and treat larval mosquito sources are critical to achieving mosquito control, especially in highly populated urban areas. The Orange County Vector Control District (OCVCD) has taken an aggressive approach to treating a variety of larval mosquito sources, especially neglected swimming pools, on private residential property through enhanced monitoring of these sources. OCVCD's program includes area-wide inspection warrants authorizing inspection and treatment of hundreds of properties in light of the recent United States Supreme Court's decision expanding Constitutional protections for areas around the home [*Florida v. Jardines* (2013) 133 S. Ct. 1409]. This presentation will discuss the Fourth Amendment's current protection of the areas surrounding the home, what constitutes a search, and how to search protected areas. The presentation will also address search warrants to inspect and treat property, including the use of "geographic-area warrants" to obtain permission to survey large areas of private property. Samples of both single property and area-wide warrants will be included with the presentation.

27 Assessing the impact of mosquito control adulticiding practices on honey bee health to improve current best management practices for mosquito control

Joseph Margotta, JMargotta@agcenter.lsu.edu, Frank Rinkevich, Brad Fritz, Clint Hoffman, Randy Vaeth, Todd Walker, James Ottea and Kristen Healy

The use of adulticiding in integrated pest management (IPM) for mosquito control is crucial in the reduction of mosquitoes as well as mosquito-borne pathogens causing human diseases, such as West Nile virus. However, the off-target effects of adulticiding practices to beneficial insects, such as honey bees in managed colonies, is largely unknown. In this experiment, we assessed acute mortality of bees at increasing distances away from a ground ULV spray truck within a 3 by 5 grid. Each station contained disposable cages containing either 20 newly emerged *Culex quinquefasciatus* mosquitoes or 20 honey bees collected from a typical colony and rotary slide spinners to measure pesticide droplet size and density. During the study we measured wind speed, temperature, and humidity, and sprayed during typical temperature inversions and wind conditions. Next, we will correlate bee mortality with droplet density, droplet size, residue, and label rates using regression analysis. These data will allow us to provide recommendations on distances to spray near managed honey bee colonies. Based on the results of this study, we will develop general guidelines for beekeepers to minimize mortality from ULV adulticide application

28 Attractiveness and impact of Terminix® All Clear® brand attractive targeted sugar bait (ATSB) to honey bees during nectar dearth

Kirk Tubbs, tfcpad@qwestoffice.net

Increases in consideration and potential use of attractive targeted sugar bait (ATSB) for mosquito abatement has raised concerns of any possible impacts on non-targeted insect foragers such as honey bees. Honey bees (*Apis mellifera*) engage in sharing food, communicate the location of food sources located at great distances from the colony, and store food for later use.

Toxic impacts can not only occur to food-foraging bees but to an entire colony with access to contaminated honey stores. Nectar is primarily composed of sugars. During times of nectar shortage honey bees search a broad area for any sugar source to use as food. They have been observed foraging for molasses on treated animal grain foods and searching around discarded soda pop containers for sugary residues. Beekeepers will feed liquid sugar solutions as a nectar replacement during times of limited food availability. This study was to gauge attractiveness and subsequent impact of ATSB (Terminix® ALL Clear® brand) in relation to natural nectars. The AI is listed as 0.40% garlic oil. The baits were put out during the period of high nectar flow and then continued through a period of (artificially) low nectar or other sugary foods availability. Bees were not attracted to the ATSB when nectar or replacement sugar solution was available. In times of nectar dearth scout bees showed some interest in ATSB placements but were not observed at repeated feedings. No toxic or other impact was noted in the hive beyond the conditions brought about by controlled food shortages.

29 *Wing Beats*: the next generation

Stephen Sickerman, sickerman@comcast.net

Wing Beats magazine premiered in 1990 as the official trade journal of the American Mosquito Control Association and the Florida Mosquito Control Association. Published quarterly, *Wing Beats* currently has a national circulation of nearly 4,000, with production and distribution costs underwritten by industry advertising. An update will be provided on editorial changes. Mosquito control professionals are encouraged

to submit manuscripts, photographs and graphics on all aspects of mosquito control, including surveillance, operations, new technologies, outreach, and environmental, legislative and regulatory issues.

30 Challenges and successes of an educational outreach program
LeShawn Simplis-Barnes, lsimplis@glacvcd.org and Andrew Pak

Vector education programs in schools are becoming more common and successful, but are not without challenges. Community participation in vector education programs is a growing practice aimed at enhancing both public knowledge and awareness of vector related issues. The Greater Los Angeles County Vector Control District (GLACVCD) has a unique mobile science lab, the VecMobile, which is available to more than 800 schools in the greater Los Angeles area and is designed to teach students about the many facets of vector biology, and the link between the environment and public health. The District's educational programs follow the California Science Content Standards and are very popular with teachers and administrators, particularly in low-income inner city areas which rarely have access to fun, educational and science-based programs for their students. The VecMobile program faces different obstacles than do traditional in-class outreach programs. The presentation will summarize more than 10 years of service and discuss the successes and challenges of marketing educational programs, budgeting, maintenance costs, program evaluation, and measuring information retention.

Chikungunya: An Emerging Threat to the Americas II

31 Chikungunya surveillance and control in New Orleans
Claudia Riegel, criegel@nola.gov and Sarah Michaels

Urban populations of *Aedes aegypti* and *Ae. albopictus,* the primary vectors of chikungunya virus (CHIKV), are widespread and abundant in New Orleans. Prolific larval habitats and substandard housing can expose residents to these vectors. There is frequent travel between New Orleans and CHIKV-infected areas of Caribbean and the return of infected travelers poses a realistic risk of establishing local transmission. In January 2014, New Orleans initiated a series of tests and procedures to reduce the possibility of locally-acquired CHIKV cases and to minimize the magnitude of an outbreak if it were to occur. In the summer of 2014, CHIKV was diagnosed in 3 individuals with travel history to the Caribbean, 1 of which was viremic upon return to New Orleans. Vector surveillance was conducted and mosquito pools were tested for the presence of CHIKV by RT-PCR. Control methods and insecticide applications were tested for efficacy. Having procedures and control measures available will strengthen the response capacity in advance of a potential domestic outbreak.

Larval Control

32 Potentials for integrated control of *Culex quinquefasciatus* (Diptera: Culicidae) using larvicides and guppies
Joy Anogwih, joyaa10@gmail.com, Winifred Makanjuola and Lucian Chukwu

Acute toxicity tests were carried out to determine the effect of 3 larvicides namely spinosad, pirimiphos methyl, and chlorpyrifos on *Culex quinquefasciatus* Say, *Anopheles gambiae s.s.* Giles (Diptera: Culicidae), and *Poecilia reticulata* Peters (Pisces: Poeciliidae). Thereafter, larvae of *Cx. quinquefasciatus* were introduced to *P. reticulata* (guppies) in containers of different volumes with low concentrations of each larvicide at established predator prey feeding densities of 1 to 35; 5 to 70 and 10 to 350 (fish to larvae), respectively. The experiment was replicated 6 times, and the population of larvae consumed by guppy was counted after 24 h. Spinosad and pirimiphos methyl were significantly ($P<0.05$) toxic to *Cx. quinquefasciatus* which was the less susceptible larval species. Guppy consumption of *Culex* larvae was highest at predator prey density 5 to 70. Feeding activity of guppy mostly increased in spinosad treatment at 49 µgL-1 when compared to control and the synthetic larvicides ($P<0.05$). The synthetic larvicides were generally characterized with a reduced rate of consumption of larvae except in the lowest treatment of pirimiphos methyl where a significant increase ($P<0.05$) in the consumption of larvae was found at the highest predator prey density in a 3 L size container. Spinosad at 49 µgL-1 with *P. reticulata* in fish to larvae feeding density of 5 to 70 contained in 0.5 L plastic produced the highest percentage population of *Culex* larvae that was consumed by the fish. Predator prey density, container size, type and concentration of larvicides are important factors for an integrated mosquito larval control system.

33 Runnelling in Australia: past present and future
Pat Dale, p.dale@griffith.edu.au and Jon Knight

This briefly overviews the history of runnelling in Australia, its more recent status and future direction. The early development of runnelling, a subset of open marsh water management (OMWM), was constrained by lack of detailed elevation data and relied on 'rough and ready' field assessment and manual labor for

construction. Later, as the method was widely adopted, mainly in subtropical Queensland, a custom built machine with laser leveler was built to construct and, where necessary, maintain runnels. Comprehensive long-term monitoring of larvae and environment was limited to only 1 site, the original Coomera Island project. Now, after several decades of runnelling, we are assessing the long-term condition of runnels and asking if they still work, how much maintenance has been involved and how we should prepare a comprehensive management plan for runnels to ensure future cost effectiveness. Finally, expanding the runnel concept of minimal modification in saltmarshes to mangrove systems is underway, facilitated by the availability of high-resolution LiDAR data and hydrological monitoring.

34 Efficacy of *Bacillus thuringiensis israelensis* (*Bti*) dunks at various temperatures against larvae of *Aedes* mosquitoes

Shabab Nasir, flourenceshabab@yahoo.com and MS Hina

Mosquitoes are vectors of many epidemic diseases such as malaria, dengue, filariasis, yellow fever and West Nile virus. Chemical control causes environmental and health hazards, so biological control is stressed. The different formulations of *Bti* are highly effective against *Anopheles*, *Aedes*, and *Culex* mosquitoes. In Pakistan, environmental conditions are very harsh; therefore we checked the efficacy of *Bti* dunks against *Aedes* larvae at different temperatures. Different concentrations of *Bti* dunks (10, 20, 50 and 100 ppm) were used against *Aedes* larvae. This research was conducted at 5 different temperatures; low temperature (10°C), medium temperature (22°C), high temperature (37°C), under shade (25°C) and under sun (30°C). The control treatments were also run against each experiment. Each trial was repeated 3 times and mortality was checked after 2, 4, 6, 12, 24, and 48 h. The results showed that efficacy of *Bti* increased with the increase in temperature due to short life cycle and frequent feeding of larvae. Life cycle of mosquito became short with increase in temperature. More mosquitoes will be produced which feed more frequently and ingest more *Bti* with food. Then ingesting more *Bti* will cause rapid deaths of larvae. But the lowest mortality was noted at low temperature (10°C) because the feeding rate of larvae became slow. Regarding concentrations, highest death rate was noted in 100 ppm and it was lowest in 10 ppm. The death rate was higher after 24 h and 48 h but it changed depending upon temperature, water grades and concentrations. It is suggested that efficacy of *Bti* can be enhanced by *Bti* dosage, *Bti* dilution, water temperature, water nature and time durations. Hence the outbreak of mosquito-borne diseases can be controlled.

35 Efficacy of *Bacillus thuringiensis israelensis* (*Bti*) dunks in different quality of water positive for larvae of *Aedes* mosquito under lab conditions

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Mosquitoes can inhabit a variety of water grades like fresh water, turbid water, foul water, sewage water etc., and act as a vector of many epidemic diseases such as malaria, dengue, yellow fever, filariasis and West Nile virus. Due to environmental and health hazards caused by chemicals, the trend is shifting towards the biological control. The use of *Bti* is highly effective against mosquitoes. In our country environmental pollution is high due to industry, inappropriate sewage system and lack of awareness. Due to this, different aquatic habitats are found, therefore we checked the efficacy of *Bti* dunks against *Aedes* larvae in different water grades under lab conditions. Different concentrations of *Bti* dunks (10, 20, 50 and 100 ppm) were used in 6 different water grades such as rich organic water, foul water, sewage water, industrial water, turbid water and useable water against *Aedes* larvae. The control treatments were also run against each experiment. Experiments in industrial water could not proceed as larvae died during acclimatization periods. The mortality of larvae was checked after 2, 4, 6, 12, 24, and 48 h. The mortality was highest in useable water, higher in foul water and high in sewage water. The low mortality was noted in turbid water and rich organic water because the organic matter, pollution and mud in water decreased the efficacy of *Bti*. The highest LC_{50} was observed in rich organic water. The statistical analysis showed that maximum LC_{50} (365.937 ppm) was observed in rich organic water under shade after 6 h. The minimum LC_{50} (2.23 ppm) was observed in useable water at low temperature after 48 h. So, the use of *Bti* is a safe and environment friendly method to combat the problem of mosquito-borne diseases.

36 Selection of *Culex pipiens* and *Culex tarsalis* laboratory strains for resistance to methoprene

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Mosquitoes are important vectors of disease and require management programs to devote vast amounts of resources. Methoprene is a tool that has high specificity and effectiveness in natural mosquito habitats where non-target populations exist. With its frequent use, methoprene is in danger of resistance development by mosquitoes. The goal of this study is to understand how the methoprene-tolerance gene (*Met*) contributes to methoprene susceptibility in *Culex tarsalis* and *Cx pipiens pipiens,* 2 West Nile virus vectors in the US. Established genomic amplification techniques (RT-PCRs) will be used to acquire *Met* gene cDNA for sequencing. The lethal dose to kill 50% (LD_{50}) of *Cx. tarsalis* (.4 mg/L) and *Cx. pipiens pipiens* (7.5 mg/L) laboratory strains using a methoprene-based commercial product (Altosid) were calculated using a survival curve bioassay. The LD_{50} was used as the starting concentration for selecting methoprene resistance in *Cx. tarsalis* and *Cx. pipiens* laboratory strains. Methoprene (Altosid) exposure in these

laboratory strains will continue through several generations. Analyses between susceptible and resistant strains will be performed to compare molecular differences. A population genetics comparison analysis is also being conducted to understand resistance in natural field populations and laboratory-selected strains of mosquitoes. Working with mosquito abatement districts throughout the state of Utah, 1,001 *Cx. pipiens* and 265 *Cx. tarsalis* 4th instar larvae were collected from 23 locations. The data collected from these laboratory studies and field collections will contribute to understanding how methoprene tolerance develops and can be predicted in *Culex* mosquitoes.

37 Mosquitoes as vehicles for insecticide

Corey Brelsfoard, cbrelsfoard@mosquitomate.com, James Mains and Stephen Dobson

Mosquito control remains the only means available to combat many medically important and nuisance mosquito species. Chemical insecticides are most commonly used to control mosquitoes, with formulations that include larvicides and adulticides. While larvicides have been proven effective at reducing mosquito-borne disease transmission, its implementation at the programmatic level can be difficult. The aquatic habitats of many mosquito species, including the Asian tiger mosquito (*Aedes albopictus*), can be small and difficult to locate and treat, sometimes referred to as 'cryptic breeding sites'. Here, we will present results of an auto dissemination approach as a method of pesticide 'self-delivery' by the mosquito to cryptic breeding sites. Laboratory and field experiments examine for model-predicted impacts of pyriproxyfen (PPF) directly applied to adult male *Ae. albopictus*, including: (1) the ability of PPF-treated males to cross-contaminate females, and to (2) directly deliver PPF to larval habitats. Data show that males contaminated both female adults and oviposition containers in field cage tests, at levels that eliminated immature survivorship. Field trials demonstrate an ability of PPF-treated males to transmit lethal doses to introduced oviposition containers, both in the presence and absence of indigenous females. A decline in the *Ae. albopictus* population was observed following the introduction of PPF-treated males, which was not observed in 2 untreated field sites. This report will also briefly summarize plans for additional work to test the ADAM approach as a mosquito control tool.

38 Pyriproxyfen: a promising insecticide for the control of Afrotropical disease vectors

Oscar Mbare, oscarmbare@gmail.com, Steve Lindsay, Bryson Ndenga and Ulrike Fillinger

Pyriproxyfen shows unique properties for mosquito control in inhibiting metamorphosis in immatures and sterilizing adults. Field studies showed that adult *Aedes* can transfer pyriproxyfen into their larval habitats to inhibit larval development. We carried out studies to evaluate these properties for control of Afrotropical disease vectors. Dose-response and semi-field tests in the open field were carried out to evaluate the susceptibility of the malaria vectors *Anopheles gambiae* and *An. arabiensis* to pyriproxyfen and determine the residual activity. Consequently, field studies were conducted to test the effectiveness of operational application of pyriproxyfen in 3-weekly intervals to natural aquatic habitats in western Kenya. In addition, exposure of adult *An. gambiae sensu stricto* and *Culex quinquefasciatus* was evaluated in laboratory at 7 time points around bloodmeal to investigate sterilization and the ability to transfer pyriproxyfen to oviposition substrates. Both *Anopheles* species were susceptible. Pyriproxyfen prevented 97% emergence of *Anopheles* larvae for a period of 6 wk in semi-field tests. Under operational application, pyriproxyfen caused 96% emergence inhibition of mosquito pupae collected from treated habitats. Females exposed to pyriproxyfen between 24 h before and after bloodmeal were 6 times less likely to lay eggs than unexposed females. Only gravid females transferred lethal doses of pyriproxyfen to cause 65%-71% emergence inhibition of larvae introduced into oviposition cups. In conclusion, pyriproxyfen presents a promising tool for integrated control of different vectors attacking their larval and adult stages and presents an opportunity for insecticide resistance management.

39 Broadcast applications of *Bacillus thuringiensis israelensis* to control tree hole breeding chikungunya and dengue vectors

James Harwood, james.harwood@med.navy.mil, Muhammad Farooq, Brent Turnwall and Alec Richardson

Aedes aegypti and *Ae. albopictus*, two of the principal vectors of chikungunya and dengue viruses, will lay eggs in water filled containers, including tree holes. Despite the risk to public health that these and other tree hole inhabiting mosquitoes present globally, there have been surprisingly few studies conducted to determine the most efficient method of applying larvicides to these habitats. Broadcast applications of 2 granular, AllPro® Sustain MGB and VectoBac® GR, and 2 liquid dispersible formulations, Aquabac® XT and VectoBac® WDG, of *Bacillus thuringiensis israelensis* from a Stihl® 450 back pack sprayer were evaluated to determine if one formulation could be better delivered to artificial containers in an open area, beneath heavy vegetation, and hung from trees, to simulate tree holes. The artificial tree holes were hung at 5 and 10 ft above the ground, at 12, 25, 35 and 51 ft from the applicator. Mortality of larval *Ae. aegypti* and droplet and granular density data were analyzed. Lethal doses of *Bti* were successfully delivered to containers up to 51 ft away, and into the artificial tree holes. Applications of granular *Bti* resulted in the highest rates of morality in all containers, regardless of how far they were from the sprayer. The different

formulations performed differently as well, with VectoBac® GR resulting in higher rates of mortality than all others.

40 Field application of VBC-60394 in a wetland habitat
Jason Hardman, jason@slcmad.org, Keith Lawson, Ary Faraji and Sam Dickson

Salt Lake City Mosquito Abatement District provides surveillance and control services for over 110 m^2 in the Greater Salt Lake City Region. However, the vast majority of control efforts are concentrated in the wetland and duck pond habitats in the northwest area of the District where large numbers of floodwater mosquitoes are produced throughout the summer. Access into these habitats is extremely difficult in the spring because of the terrain and environmental sensitivity where large varieties of bird species feed and nest. Additionally, because seasonal staff are not hired until the beginning of summer, lack of available personnel, economics, logistics, and time constraints further exacerbate much needed resources during the early season. In order to circumnavigate some of the above issues, we evaluated a pre-hatch application of an experimental mosquito control pesticide (VBC-60934) to be used in floodwater habitats of our District. The product was easily applied using standard larval control equipment, was efficacious, cost effective, did not break down under climatic conditions, and was environmentally sound. Our presentation will highlight the trials and tribulations of this product and provide insight into the operational use of this pesticide in our habitats and regions.

41 The joy of controlling *Aedes dorsalis* on a small, coastal Oregon salt marsh
Daniel Markowski, dmarkowski@vdci.net and William Johnson

Beginning in the fall of 2012, an extraordinary mosquito problem stemming from a marsh reclamation project began brewing in a small, coastal Oregon town. The conditions of the marsh coupled with the tides and mild summer weather created unprecedented nuisance levels for the nearby community. Large populations of *Aedes dorsalis* were affecting the quality of life for neighboring citizens, recreation of both citizens and visitors to the area, and occupational health of those working outdoors in the affected area. In 2013, adult light traps on the refuge collected nearly 5,000 mosquitoes in a 12-h period while larval surveillance resulted in dips producing as many as 300 larvae per dip. US Fish and Wildlife Service (USFWS) implemented a mosquito identification and monitoring plan in the spring of 2013 and ultimately a mosquito control plan for the marsh in the summer of 2014. Larvicide treatments began in late April and continued throughout the summer of 2014. In May, 132 acres were treated after the full moon tide. In June and July aerial applications were conducted. By the September high tide, the acreage that needed treatment was greatly reduced through ditching efforts, begun by the USFWS in late July, with only 31 acres needing treatment. Existing within the intersection of politics, science, operational realities, and environmental concerns, this project tested our resolve in many ways that will be discussed.

42 Plant-based isoflavones show effective larvicidal activity with low mammalian toxicity
Jeremiah Malerich, jeremiah.malerich@sri.com, Mary Tanga and Alden Estep

The mosquito *Aedes aegypti* is the principal global vector of the viruses that cause dengue fever, yellow fever, and chikungunya. Environmentally safe strategies that exploit novel modes of action to control mosquitoes are desperately needed. Flavonoids are polyphenolic plant compounds that have shown promise as mosquito larvicidal agents, although little is known about their modes of action. We found that 100 µM of the flavonoid glycoside rutin controlled >50% of *Ae. aegypti* 1st instar larvae in 24 h. Encouraged by this result, we screened a series of rutin derivatives and identified a water-soluble, chlorinated flavonoid with an EC_{50} value of 4.5 µM that did not exhibit cytotoxicity against a human kidney cell line at 1-10 µM. Halogenated rutin derivatives exhibit low vertebrate toxicity, and studies indicate that these molecules kill insects by a mode of action different from existing insect growth regulators and microbial gut toxins. We conclude that rutin derivatives are rational candidates to develop environmentally advanced larvicides.

Research Highlights 2015

43 Highlights of tick biology
Lorenza Beati, lorenzabeati@georgiasouthern.edu

A rapid search through tick-related literature reveals an abundance of reports about tick-borne pathogens and their detection in ticks. Although these topics are certainly of great importance in terms of public health, in this presentation we will focus on other aspects of tick biology. In this past year, several studies have changed what we thought we knew about some very common tick species. From the fundamental questions on tick taxonomy and evolutionary history, to ecology and control, we will explore the 2014 tick literature by emphasizing publications which made us revisit some well-established certitudes.

44 Highlights of mosquito biology
Ary Faraji, ary@slcmad.org

This presentation will summarize recent published reports in the scientific literature describing the latest advancements in mosquito biology and ecology. Special emphasis will be given to invasive mosquitoes and the challenges they pose in regards to public health and comfort. A selected number of publications will be chosen and discussed, with the aim of providing the audience an inclusive summary between the academic and operational perspectives.

45 Highlights of mosquito control: New technologies for the control of mosquitoes--parasites, gene manipulation and lasers!
Derric Nimmo, derric.nimmo@oxitec.com

First there was *Aedes aegypti* that spread across the world from Africa then *Ae. albopictus* followed from Asia. Soon after came dengue and now there is chikungunya spreading rapidly throughout the Caribbean in only a matter of months! Vector control has never been so advanced but the spread of these mosquitoes and the diseases they carry has not been stopped. New methods are desperately needed to help combat these mosquitoes. There are several promising new technologies out there, including *Wolbachia*, the sterile insect technique (SIT), transgenic technologies to enhance the SIT and gene drive systems to drive resistance genes through a population. Each of these technologies has its challenges and pros and cons, and some are more advanced towards field application than others. However expect to see some of these technologies available to mosquito control districts within the next couple of years! This talk will review these new technologies and talk about what's just around the corner to help combat *Aedes* and other mosquito species.

Student Competition I

46 The 26th student paper competition of the American Mosquito Control Association
Brian Byrd, bdbyrd@wcu.edu

Since 1989 the American Mosquito Control Association has encouraged student participation by providing student members a forum to present their research. This venue is judged competitively and winners receive the Hollandsworth Prize, which honors AMCA member Gerald Hollandsworth. Each participating student will have 15 minutes to present their paper.

47 Subacute exposure to prallethrin modifies behavior of medically important vectors
Kyndall Dye, kyndall.dye@uky.edu, Grayson Brown and Douglas Johnson

A randomized complete block design was used in woodlot edges to test the locomotor effects of prallethrin, a new, yet costly, addition to ULV mosquito adulticides, on sand flies, ticks, and mosquitoes. No significant effects on trap catches of any vectors were detected. A laboratory spray system was constructed simulating ULV application to mosquitoes which showed locomotor stimulation with direct contact. This helps explain the anecdotal price performance reports of adulticides containing prallethrin.

48 Impact of application site and adult age on the efficacy of two topically-applied insecticides to *Culex quinquefasciatus* Say
Robert L. Aldridge, robert.aldridge@ars.usda.gov, Phillip E. Kaufman, Jeffrey R. Bloomquist, Salvador A. Gezan and Kenneth J. Linthicum

In order to determine if age and application site contribute to insecticide susceptibility, precision droplet applications of permethrin and malathion were topically administered to unconventional body regions of mosquitoes across several adult age classes. Our results indicate that droplet application site influences susceptibility to these insecticides. Furthermore, insecticide bioassay procedures may require modification to reflect these age and morphological susceptibility results, potentially reducing insecticide use through targeted applications.

49 Impacts of garlic formulation in ATSB on feeding and mortality of three mosquito species
Rebecca Zimler, razimler@ufl.edu

Attractive Toxic Sugar Bait (ATSB) is a new approach of mosquito control with increasing interest in using alternative active ingredients. One of these botanical active ingredients is garlic, a known mosquito repellent. In this study, we report on feeding behaviors and mortality of three mosquito species (*Aedes aegypti*, *Culex quinquefasciatus*, and *Anopheles albimanus*) given different concentrations of ATSB containing garlic. Excreted droplets containing FD&C Blue #2 dye provided an index of consumption of ATSB formulations.

50 Insecticide incorporated mesh barrier for host-seeking biting midge (*Culicoides sonorensis*) control

Darren Snyder, snyderd@ksu.edu and Lee W. Cohnstaedt

Impeding potential pathogen carrying biting midges from reaching their host is a daunting task. Treated barriers around livestock can be used to control the number of biting midges reaching a host, attaining a blood meal and possibly transmitting a pathogen. Addressed here is the use of an insecticide incorporated barrier to reduce the number of host-seeking midges able to enter a mock animal enclosure surrounded by the treated material.

51 A melting curve PCR assay that can detect the *ace*-1 mutation in *Culex quinquefasciatus*

Cassie Scott, gni6@cdc.gov

Insecticide resistance is a threat to public health. Acetylcholinesterase resistance is due to mutations in the *ace*-1 gene that decrease organophosphate sensitivity. A melting curve assay was designed to detect the glycine-serine *ace*-1 mutation in *Culex quinquefasciatus*. Allele-specific PCR was used to detect single nucleotide polymorphisms. Three different melting curve profiles were observed indicating homozygous and heterozygous genotypes. This tool may be used for early resistance detection, which is critical for development of management strategies.

Adult Control I

52 Insecticide resistance management: the Zambian experience

Dayton Makusa, day_t_on@yahoo.com and Chadwick. H. Sikaala

Resistance against insecticides is an increasing challenge and a major threat to malaria vector control in many African countries including Zambia. Given that there are a limited number of insecticide classes available for mosquito control (i.e., organophosphates, organochlorines, carbamates and pyrethroids), selection pressure is exerted, thus vector populations increase drastically. The necessity of having an effective long-term resistance management cannot therefore be overemphasized. However, many factors require consideration to achieve this, going beyond mere availability of insecticides, training of vector control personnel to ensure proper implementation and surveillance and availability of educational materials may be critical.

In view of this, the Zambia National Malaria program (NMCP), with support from USAID (PMI) through ZISSP/AIRS project, has put in place a strategic institutional framework that brings together the NMCC and other malaria research institutions that include the Macha Malaria Research Institute, Tropical Diseases Research Centre, The Liverpool School of Tropical Medicine and the Malaria Transmission Consortium to take up the challenge of leading the insecticide resistance management activities through the Resistance Advisory Committee and the Insecticide Resistance Management Technical Working group.

The work from the resistance management technical working group has been used to inform policy and has since been the basis for selection of insecticides for use in indoor residual spraying activities for the past 4 yr. This has been in combination with the training given to Environmental Health staff in basic entomology, spraying technique and volunteer spray operators training.

53 Mosquito and filth fly control in desert and temperate environments with a synergized pesticide mister and barrier treatment

Seth C. Britch, seth.britch@ars.usda.gov, Kenneth J. Linthicum, Robert L. Aldridge, Jerry D. Kerce, Jeremy Wittie, Gregory White, Melissa Snelling and Arturo Gutierrez

US military operations face significant negative impacts on mission readiness from disease-vector and nuisance filth flies, mosquitoes, and sand flies. Through the Deployed War Fighter Protection Program (DWFP) we previously developed small scale 9 X 3 ft pesticide-treated perimeters enhanced with timed pesticide misting systems that reduced populations of these insects or reduced insect-human contact in a range of environments. In this study we investigated large scale 42 X 39 ft treated HESCO blast wall perimeters enhanced with a pesticide misting system. We conducted preliminary trials using a permethrin formulation against mosquitoes in a warm humid environment in north-central Florida, and set the mister to spray outwards from the top of the perimeter during peak night mosquito activity. Although population numbers were low overall, mosquito numbers were somewhat reduced within the large HESCO perimeter with the mister present. Later more extensive trials were conducted with a similar perimeter and mister configuration against filth flies in a hot-arid desert environment in southern California during peak morning fly activity. Efficacy was measured by the number of flies captured in 20 min replicates within the perimeter lacking the mister compared to the number captured during 20 min replicates of pulsed misting with the permethrin formulation. When the spray was directed towards the outside of the perimeter, flies were reduced by >71%; however, when the spray was directed towards the interior, the reduction was >84%.

We discuss potential real-world military and civilian applications of this configuration to reduce nuisance flies and transmission of enteric disease pathogens.

54 Can adult mosquito traps play a larger role in a dengue/chikungunya response plan?
Aaron Lloyd, alloyd@pascomosquito.org, Mike Greer and Dan Kline

The Asian tiger mosquito, *Aedes albopictus* (Skuse), ranks among the most significant vectors of dengue fever and chikungunya virus. With no specific medications or vaccines available, vector control is the only way to combat these diseases. Public health programs concentrate on source reduction, public education and in some cases adulticiding when responding to a virus introduction. Mosquito traps are commonly used for surveillance but may be able to play a larger role in helping to reduce the adult mosquito population when there is a disease threat. The objective of this study was to investigate the use of BGS traps to quickly reduce adult Asian tiger mosquito populations in an established tire pile in Pasco County, FL. Results from this study and plans for future studies will be discussed.

55 Chikunguyna and dengue vector control strategies: evaluation of two wide-area mosquito control strategies targeting *Aedes aegypti* and *Ae. albopictus* populations
Christopher Lesser, Christopher.Lesser@manateemosquito.com

The Manatee County Mosquito Control District (FL) evaluated and compared the population control efficacy of multiple night-time aerial-applications of Fyfanon (AI 97% malathion, Cheminova) vs. efficacy of multiple aerial adulticide + larvicide application (Fyfanon + Altosid 5%). Mosquito population dynamics were measured via landing rate counts over 3-mo period. Spray cloud dynamics were measured at each study site. Mosquito populations subjected to aerial adulticide (only) dropped significantly (97.2 to 100%) immediately after individual treatments but rebounded to pre-treatment levels within 1 wk; net population reduction over the study period was 65.5% as compared to pre-treatment and control population trends. Comparatively, mosquito populations subjected to both larvicide and adulticide treatments in a surveillance-based IPM approach observed an 89.3% reduction over the course of a 3-mo period as well as a similarly fast knock-down of adult mosquito populations immediately after each aerial adulticide mission (85.2 to 93.0%). Results of this study indicate that multiple aerial adulticide missions can be very effective in quickly reducing large domestic mosquito populations as may be needed during a dengue or chikungunya disease epidemic. Alternatively, if long-term and wide-area population reduction is desired, surveillance-based larvicide applications are a necessary addition to aerial adulticiding.

56 Botanical oils enhance potency of synthetic pyrethroids as well as or better than piperonyl butoxide (PBO)
Joel Coats, jcoats@iastate.edu, Lyric Bartholomay, Aaron Gross and Edmund Norris

Infectious diseases spread by mosquitoes are significant problems in many locations throughout the world. The yellow fever mosquito (*Aedes aegypti*) is the primary vector of the dengue virus, a rapidly spreading mosquito-borne virus. *Anopheles gambiae* is a vector of the malarial *Plasmodium* parasites. Chemical control of mosquitoes has been hampered by: 1) increasing insecticide resistance, and 2) increased public concern about the safety of chemical insecticides. Use of chemical insecticides with synergists, like piperonyl butoxide (PBO), has been partially effective at overcoming resistance and reducing amounts of insecticides used. Terpenoids in plant essential oils have been shown to be mildly effective in killing mosquitoes. In addition, essential oils and their terpenoids can enhance the potency of natural pyrethrins. In an effort to identify plant essential oils that could be utilized to enhance the activity of pyrethroids, 30 plant essential oils, from various plant families, were topically applied to adult female *Ae. aegypti* and *An. gambiae*, to determine their baseline toxicity. In addition, mixtures of plant essential oils and a commercially available permethrin formulation were analyzed for enhanced efficacy and compared to PBO. Numerous plant essential oils are as effective via topical application as PBO, or more effective as enhancers when compared to PBO. Several oils are also comparable in mixtures with permethrin or deltamethrin in WHO cone tests (contact-toxicity tests). Aerosol formulations have been generated by our commercial partner EcoSMART Technologies, Inc., Roswell, GA.

57 Withdrawn

58 Assessing the effects of mosquito control spraying on butterfly populations
Marc Minno, marc.minno@gmail.com

Four butterfly species in southern Florida are now listed by the US Fish and Wildlife Service as endangered and several others are under review. Application of mosquito adulticides is frequently blamed as a major cause of population declines in non-target invertebrates such as butterflies. For decades, the Florida Keys Mosquito Control District has maintained an active program to control mosquitoes that includes spraying adulticides from trucks and aircraft.

To determine patterns of species richness and distribution, I searched for butterflies from August 2006 through December 2012 throughout the Florida Keys in monthly to bimonthly surveys. I found 69 species of

butterflies and tallied 43,381 adults in more than 1,150 h of observation on 42 islands (1,381 site visits). Fewer butterflies were found in the Upper and Middle Keys (53 species, 11,308 adults) compared to the Lower Keys (61 species, 32,073 adults).

Butterflies have disappeared from vast areas of southern Florida that are not sprayed such as the Ten Thousand Island National Wildlife Refuge, Everglades National Park, and Biscayne National Park. Some urban parks on Key West, Stock Island, and a private butterfly garden on Big Pine Key were among the places that had the greatest abundance and diversity of butterflies in the Keys, while many public conservation lands had relatively few butterflies.

The Cassius blue, giant swallowtail, zebra heliconian, and Gulf fritillary were among the most abundant and widely distributed butterflies in southern Florida. Yet these species have been shown to be highly sensitive to mosquito adulticides in laboratory studies. The major cause of population declines of butterflies in southern Florida is likely due to exotic predatory ants.

59 An autocidal *Wolbachia*-based approach against *Aedes albopictus*

James Mains, jmains@mosquitomate.com, Corey Brelsfoard and Stephen Dobson

The Asian tiger mosquito (*Aedes albopictus*) is an invasive species and public health concern due to its ability to transmit medically important pathogens (e.g., dengue, chikungunya) and aggressive day-biting behavior. Despite intensive use of pesticides to manage this species, it has colonized much of the USA, and in recent years its range has expanded to include California and New York. A proposed autocidal approach for its control is based on *Wolbachia*, an endosymbiotic bacteria that is common in many insect species. Similar to sterile insect technique, the *Wolbachia* approach is based on the release of *Wolbachia* infected males, which cause a form of conditional sterility in the targeted populations. On July 26, 2013, MosquitoMate Inc. was awarded an EPA Experimental Use Permit (No. 89668-EUP-1) to conduct field performance testing of this approach for the Asian tiger mosquito in the continental US. Presented here, are early results of preliminary, open release field trials to examine for *Wolbachia* infected male *Ae. albopictus* performance under field conditions.

60 Vector-control with garlic-oil: a new recipe of attractive toxic sugar bait (ATSB) significantly diminishes the *Anopheles sergentii* population in a desert oasis

Günter C. Müller, guntercmuller@hotmail.com, Yosef Schlein and Edita E. Revay

Attractive toxic sugar bait (ATSB) is a highly effective vector-control method which targets mosquitoes based on their foraging behavior, by applying insecticide solutions to plant species known to be important sugar-sources of local mosquito populations, or by combining oral toxins and sugar with known attractive compounds. Environmental and selection-pressure concerns have prompted investigations of novel, ecologically-harmless substances which can be used as insecticides. This study utilized microencapsulated garlic-oil as the oral toxin component of ATSB, which was found to be an effective insecticide for controlling *Anopheles sergentii* populations inhabiting desert-surrounded wetlands in Israel. Biting-pressure values in the ATSB treatment site decreased by 97.5% over the experimental period, while at the control site treated with non-toxic attractive sugar bait (ASB), no significant changes were observed. Only 18.5% of the non-target insects were found to have ingested ASB-derived sugar prior to capture, of which 50% were non-biting midges of the order Diptera. While non-biting midges ingested bait-derived sugars applied to both flowering (75%) and non-flowering plants (25%), other non-target insects were found to have foraged almost exclusively on ASB applied to flowering-plants (93%), with only 7% found to have ingested foliar-applied bait. These results demonstrate microencapsulated garlic-oil can be used as an alternative to traditional pesticides, with minimal environmental impact when applied in accordance with label recommendations.

Extension Medical Entomology Programs: Current Programs and Future Challenges

61 Incorporating technology into extension programs

Kristen Healy, khealy@lsu.edu

Technology is advancing at a rapid pace, as smartphones and computers become common in most people's homes. According to several popular marketing websites, it is estimated that 60% of the population has a smart phone or cell phone that is used to access the internet. According to many faculty and administrators at universities, this rapid adoption of technology has clearly changed how students are learning. In a recent survey of our target audience for a new extension program, we found that the majority of survey respondents were more interested in taking online modules over traditional in classroom learning. Therefore, as educators involved in extension and teaching, it is essential to not only keep up with technology, but to find ways to adopt it into our programs. This may include development of cell phone applications, distance education, developing online interactive curricula, or working with graphic designers to develop fun educational materials. An advantage to these types of tools is that they are low cost and far-

reaching. While technology might seem intimidating to some users, recent programs have been developed to help users create material without the need for understanding programming language. Therefore, numerous opportunities exist to incorporate technology into extension programs.

62 Maintaining relevancy in the 21st century

Elmer Gray, ewgray@uga.edu

The University of Georgia has an active and diverse Department of Entomology with 36 full-time faculty members. Eleven of these faculty members and 2 staff members have full or partial extension appointments. Growing up on the coast of New Jersey, I experienced the mass emergences of mosquitoes that can occur in the coastal environment. It is the annoyance, aggravation, and vector potential of large mosquito populations that will help us maintain our relevance in the 21st century. While mosquito-borne diseases are extremely serious, their occurrence is often highly localized and sporadic. Conversely, when environmental conditions create large mosquito populations, large areas and populations can be affected and often the public will demand action whether disease transmission is a threat or not.

The action usually required is an adulticide application. However, adult mosquito control is a very visible and occasionally controversial activity. As a result, it is extremely critical that mosquito control operators be highly trained in all aspects of mosquito biology, surveillance and control. Public confidence in the competence and professionalism of mosquito control operators is essential in today's society. The role of training mosquito control operators is well suited to those of us in the Extension Medical Entomology field. Properly trained mosquito control technicians will be more efficient and effective and more likely to conduct their work according to integrated pest management principles. The future of the profession depends on properly trained professionals and the cooperative extension service will be essential in this endeavor.

63 Extending knowledge and changing lives

Rebecca Trout Fryxell, RFryxell@utk.edu

La Crosse encephalitis is the leading arboviral disease among children, and was previously limited to the Upper Midwest. In 2012, 9 pediatric cases of La Crosse encephalitis occurred in eastern Tennessee, including 1 fatal case. In an attempt to identify potential field sites for future La Crosse virus (LACv) mosquito studies near the fatal case, the University of Tennessee Institute of Agriculture county extension offices were contacted. With their help, we initiated an end of season study which included 49 sites within 16 radial km of 2 pediatric infections. LACv was isolated from 3 *Aedes triseriatus* pools and the generated genome sequences were 100% identical with the autopsied case. Collaborations with Extension are indispensable; methods of sneaking Extension into a research program will be discussed.

64 West Nile virus outbreak and the effects it had on extension medical entomology in Texas

Mike Merchant, m-merchant@tamu.edu, Gabriel Hamer and Molly Keck

The first case of West Nile virus occurred in Texas in 2002 but prior to the WNV epidemic of 2012, few cities and municipalities conducted mosquito surveillance or had employees dedicated for mosquito management. For several years prior to the outbreak, medical entomology positions at the state health service and agricultural Extension service were mostly vacant. Most funding previously used for mosquito management throughout these entities was obsolete.

During the 2012 outbreak, municipalities scrambled to train staff in the basics of mosquito management, assemble a mosquito monitoring program and develop appropriate management plans. Cities were racing against nature to stop the spread of West Nile virus. Extension entomologists and state and county health services were being called continually for help, support and information. In addition, the CDC was asked to assist with evaluating the epidemic response and planning for future WNV emergencies.

After the outbreak, the Texas A&M AgriLife Extension Service and the Department of Entomology at Texas A&M University organized a statewide task force to better serve the state health services, cities, municipalities and the public. Educational programs were developed and administered and new personnel were hired throughout the state. The overall impact from the 2012 outbreak was a major learning curve and wake-up call for Texas.

65 Extension medical entomology as a research tool: a win-win

Dina Fonseca, dinafons@rci.rutgers.edu

From 2008 through 2012, we developed a multidisciplinary area-wide approach grounded by an economic analysis to control the Asian tiger mosquito, *Aedes albopictus*, an important nuisance and disease vector in the US. Our current guidelines incorporate passive and active public education with area-wide application of biological and chemical control interventions informed by a degree-day approach. The basis for the project's success was the combination of operational research in biology, economics, and education grounded by the humbling need to show results in intervention campaigns directly engaging with homeowners in high density residential areas. These actions were only possible with the full partnership of 2 county mosquito control

programs in NJ. The results of the operational research improved our understanding of the sources, physiology and spatial architecture of US populations of *Ae. albopictus* allowing customization, better ways to achieve homeowner involvement and a better understanding of the added value of mosquito control. A win-win.

66 USDA-ARS extension activities in medical, veterinary and urban entomology
Kenneth Linthicum, kenneth.linthicum@ars.usda.gov

Within the USDA Agricultural Research Service (USDA-ARS), National Program 104 conducts research on veterinary, medical, and urban entomology. The goal of this program is to develop more effective methods of preventing or suppressing insects, ticks, and mites that affect animal and human well-being. Research is focused on sensory ecology, problems associated with higher flies, geographic information systems and spatial analysis, and invasive species. Each of these research areas represents a group of interests that cut across the efforts of multiple research management units. The research of each of these units, located across the US, will be described. Technology transfer of research results and new products are a key activity to support key stakeholders and ultimately to improve the health and well-being of humans and animals. Linkages with partner federal agencies such as other USDA-ARS National Program Areas, the USDA-Cooperative State Research, Education, and Extension Service, the Environmental Protection Agency and the Department of Defense enhance and improve the ability of National Program 104 to solve agricultural problems associated with many kinds of pests.

Looking In Our Rear View Mirror: Events, Personalities and Discoveries That Have Shaped Mosquito Control I

67 Arboviruses and mosquito control "down under": Are we on top of it?
Scott Ritchie, scott.ritchie@jcu.edu.au

Australia straddles the Tropic of Capricorn, and features mosquito/arbovirus ecosystems ranging from temperate to subtropical and tropical. The so called Lucky Country has been plagued by periodic epidemics of several arboviruses causing disease and even fatalities in man. Epidemics of the potentially fatal flavivirus Murray Valley encephalitis virus led to the establishment of sentinel chicken based surveillance programs in southern Australia. In the 1970s-80s, the alphavirus Ross River virus was recognized as causing annual outbreaks of 1,000s of cases of epidemic polyarthritis. The linking of RRV to several species of common mosquitoes led to expansion of mosquito control programs in cities such as Brisbane and Perth that had annual outbreaks of this disease. Dengue re-emerged in 1981, and now annual outbreaks occur in north Queensland. Finally, globalization has led to increased importation of exotic vectors and pathogens. The Indo-Papuan conduit that connects northeastern Australia to Indonesia via Papua New Guinea and the Torres Strait has been a bridge over which exotic vectors (*Aedes albopictus, Culex gelidus*) and viruses (dengue, Japanese encephalitis) have been introduced into Australia. Currently, *Ae. albopictus* threatens to invade the Australian mainland, and chikungunya and Zika viruses, both of which are active in the Western Pacific, could exploit this opportunity. We are in a rapidly changing world, and both mosquitoes and the pathogens they carry offer a dynamic threat to Australia. New technologies, such as *Wolbachia*, coupled with boots-on-the-ground mosquito control, offer the potential to keep Australia free from the worst that these threats pose.

68 Rediscovering the Yellow Fever Commission: A personal journey
Natasha Agramonte, nme@epi.ufl.edu

I trace the life and public health contributions of Aristides Agramonte, the Cuban-American member of the Yellow Fever Commission of 1901. The Yellow Fever Commission was led by Walter Reed, with assistance from James Carroll, Aristides Agramonte, and Jesse Lazear. Their efforts led to the discovery that the yellow fever virus was transmitted by a mosquito, *Aedes aegypti*. Little is publicly known of what happened to Agramonte after his work with Walter Reed. My own connections to this story are also discussed.

69 Bridge on the River Kwai: the man, the movie
Joseph Conlon, conlonamcata@gmail.com

A winner of 7 Academy Awards, the movie, "Bridge on the River Kwai" is a fictionalized depiction of actual events on the so-called "Death Railway" from Bangkok to Rangoon during the Second World War. The bridge in question was actually built by Allied prisoners of war led by a LtCol Phillip Tousey, played in the film as LtCol Nicholson by Sir Alec Guinness. The presentation will explore the manner in which tropical diseases affected the prisoners constructing the bridge in addition to discussing in detail the differences between the actual events and their depiction on screen. It will further bring to light the efforts of LtCol Tousey, after the war, to address the myriad disease sequelae suffered by his troops through his association with the Liverpool School of Tropical Medicine.

Student Competition II

70 **Activity patterns of *Aedes albopictus* within a diverse environment of residential and agricultural activity and introduction of a new lethal ovitrap for controlling wild vector populations**

Casey Parker, caseyparker@ufl.edu, Alexandra Chaskopoulou, Emmanuel Fotakis, Roberto Pereira and Philip Koehler

These studies determined the optimal location and timing for lethal ovitrap deployment and maximum efficacy. Studies on the distribution of *Aedes albopictus* in coastal Greece showed tires, drainage systems, and containers as primary breeding sites. Adult surveillance using CDC light trap, BG Sentinel traps, and standard ovitraps indicated population peaks in late July-early August. Durable dual-action lethal ovitraps (DDALO) caused ~98% mortality in lab studies. The combination of field and lab studies supports DDALOs deployment for controlling mosquito populations.

71 **Design and synthesis of new α-aminonitriles via Strecker reaction. Search of molecules with insecticide action and its effect on the mosquito *Aedes* (*Stegomyia) aegypti*, vector of dengue and chikungunya fever**

Andrés Rueda, gilberto918@hotmail.com, Aurora Carreño O, Jonny E Duque and Vladimir V Kouznetsov

Efforts to controlling disease vectors of dengue and Chikungunya fever are compromised due to the resistance developed by mosquitoes to commercial insecticides. The search and discovery of molecules capable of inhibiting the enzyme acetylcholinesterase (AChE) has outlined new insecticides, which is a priority due to the lack of vaccines to prevent or drugs to treat these mosquito-borne diseases. A new series of nine girgensohnine analogs (moderate AChE inhibitors) were synthesized by a Strecker reaction modification, catalyzed by sulfuric acid supported on silica gel (SSA). By computational methods, the ADMETox parameters were obtained. Structural confirmation was determined by physicochemical, spectroscopic and spectrometric methods. Larvae between third and fourth instar and adults of a Rockefeller colony of *Ae. aegypti* were used to determine the larvicide and adulticide activities of the analogs and LC_{50} and LC_{98} values were calculated by probit analysis. Exploratory doses used in the larvicidal activity were 50, 70 and 120 ppm whereas for adulticide activity concentrations of 1000, 300 and 30 ppm were evaluated. Analogs showed larvicidal activity below 150 ppm for all the synthetized molecules. The molecule 2-(4-methoxiphenyl)-2-(4-methylpiperidin-1-yl) acetonitrile showed the highest larvicidal activity between the nine molecules evaluated, with an LC_{50} = 69.58 ppm and LC_{98} =139.54 ppm. All synthesized compounds were characterized and their molecular descriptors suggest its biological application without any theoretical toxicity risk. The analogs presented larvicidal and adulticide action on *Ae. aegypti* larvae and mosquitos respectively, under laboratory conditions.

72 **The effects of foliage diversity to applications of Attractive Toxic Sugar Bait on non-target arthropods**

Jodi Scott, bowerj@ufl.edu

The correlation between foliage diversity and non-target arthropods was assessed through simulated Attractive Toxic Sugar Bait treatments. Dyed Attractive Sugar Bait (no active ingredient) was applied to two 50 m sites each comprised of different foliage diversity. Treatments were assessed by multiple collection devices up to 24 h post application. The site with greater foliage diversity had the highest number of arthropods collected while the lowest number that consumed the bait (3%).

73 **What is the purpose of multiple lobed spermathecae in *Aedes aegypti* and *Aedes albopictus*?**

Carrie DeJesus, cedejes2@ncsu.edu and Michael Reiskind

Aedes aegypti and *Ae. albopictus* are two important disease vectors of chikungunya and dengue. These two species share many life history traits: geographic distribution, container breeders, and mate near their host. They also share similar physiologies including a three lobed spermathecae. There is one large lobe (medial) and two smaller lobes on each side. Typically sperm is found within the medial lobe with little to no sperm within the lateral when intraspecific mating occurs. *Aedes albopictus* has become a successful invasive species and has extirpated populations of *Ae. aegypti* in the United States, possibly through competitive displacement of *Ae. aegypti* by *Ae. albopictus* by satyrization. This occurs when *Ae. albopictus* males successfully mate with *Ae. aegypti* females rendering *Ae. aegypti* females sterile. Little is known about the mating behaviors of these species. Further investigation into the roles of the spermathecae could provide insight on the mating biology of these two critical disease vectors. In this investigation, I examined how body size and mixed matings may impact the filling of spermathecae in both *Ae. aegypti* and *Ae. albopictus*. I investigated the effects of body size, time of mating, and interspecific mating. I hypothesize that females will fill more lobes and take less time with large males of conspecifics compared to small males. I hypothesize that interspecific matings *Ae. aegypti* will fill more spermathecal lobes interspecific matings than *Ae.albopictus*. Results and implications of this study are discussed.

74 Sterilization with no evidence of insemination of *Aedes aegypti* by invasive *Aedes albopictus*
María Cristina Carrasquilla, mccarrasquilla@ufl.edu

Satyrization is the probable cause of competitive displacements of *Aedes aegypti* by invasive *Aedes albopictus*. Although interspecific mating rates are low in nature, as measured by heterospecific sperm in spermathecae, on the basis of cage experiments using stable isotopes and refractoriness to subsequent conspecific mating, we demonstrate that *Ae. albopictus* males may sterilize *Ae. aegypti* females without sperm transfer to spermathecae. Thus, heterospecific sperm detection in wild-caught females may underestimate satyrization effects in nature.

75 Determining optimal source material of oviposition attractants from the larval rearing medium of *Phlebotomus papatasi*, the vector of old-world zoonotic cutaneous leishmaniasis
Bahjat Fadi Marayati, b_maraya@uncg.edu, Charles Apperson, Coby Schal, Loganathan Ponnusamy, Madhavi Kakumanu and Gideon Wasserberg

The control of sand flies, the vectors of the *Leishmania* parasite worldwide, is currently limited to personal protection and residual spraying. Therefore, targeting control efforts on gravid females could affect disease transmission by reducing the abundance of blood-fed females and by suppressing the vector's population growth rate. The study presented here describes results from a broader project in which we apply a systematic integrated approach including behavioral, electro-physiological, and microbiological investigations to elucidate the cues that drive the oviposition behavior of the sand fly *Phlebtomus papatasi*, the vector of old-world Zoonotic Cutaneous Leishmaniasis. We have used a multiple-choice oviposition bioassay in which gravid *P. papatasi* females were presented with five different source materials from their rearing medium on filter paper disks in oviposition cups. We also evaluated whether this oviposition response changes with female's age. The top three candidates were tested individually in two-choice oviposition cup bioassays. Our results suggest that the oviposition rate was highest on the filter paper with the 2nd-3rd instar larval substrate, followed by the 4th instar larval substrate, rabbit feces, and larval food. To evaluate whether the total oviposition response measured in the oviposition cup bioassays also indicates attraction, we conducted olfactometer bioassays using a 3-chamber linear olfactometer in which we tested all five candidate materials. Our results confirm the trend from the multiple-choice experiments with the substrate extracted from the 2nd-3rd instar being the most attractive followed by the 4th instar larval substrate and the rabbit feces.

Adult Control II

76 Modeling biotic and abiotic drivers of public health risk from West Nile virus in Ohio, 2002-2006
Paul Rosile, MPH, PhD, paul.rosile@eku.edu, Song Liang, PhD

The objective of the study was to develop practical predictive models to be used in mosquito control programs. Weekly temperature (T), weekly precipitation (CP), and the Palmer Index (PDI) were used to inform time-delayed indices of the mosquito life cycle. Descriptive statistics were used to characterize temporal and spatial patterns of human WNV disease, WNV positive birds, *Culex* spp. infection rates (IRs), and mosquito density. Regression analyses were performed to examine the meteorological drivers underlying WNV infections in mosquitoes. A mathematical model (MM) was developed, evaluated, and calibrated using actual mosquito infection data.

In 2002, the human case onset epidemic peak was preceded by 1 to 2 wk by mosquito infection rate peaks, 3 to 4 wk by peaks in bird deaths, and 5 to 6 wk by peak mosquito density. Increases in IRs were associated with an increase in T during oviposition, the gonotrophic cycle, egg-to-hatch (E-H), during the 6 wk prior to E-H, the overwintering time-delayed indices, and changes in CP and the PDI. The MM that produced the best control strategy was a combination of 10% larvae and 30% adult control. Increasing the weekly bite rate and decreasing the mosquito natural mortality rate changed the timing and magnitude of peak infected mosquito density. Mosquito death rate was adjusted from 0.59 to 0.70 per mosquito per week to attain a $R0 < 1$. Control strategies were adjusted to a 12% weekly larval reduction combined with a 5% weekly adult reduction to produce an estimated $R0 < 1$.

The public health implications of this study should be realized with continued research on connecting the mathematical model outcomes to real-life applications of mosquito vector control.

77 Pyrethroid resistance status of *Aedes aegypti* (Diptera: Culicidae) in Thailand
Waraporn Juntarajumnong, waraporn2521@gmail.com, Sungsit Sungvornyothin, Chanita Kantaniyom, Patcharawan Sirisopa and Theeraphap Chareonviriyaphap

Aedes aegypti is a primary vector for dengue fever and dengue hemorrhagic fever worldwide. Prevention and control of the disease are almost dependent on vector control using synthetic pyrethroids. This could be

an important cause of insecticide resistance. Therefore, *Ae. aegypti* from 8 localities in northeastern, southern and western with a current dengue outbreak in Thailand were subjected to 6 synthetic pyrethroid insecticide susceptibility assays. The results revealed that *Ae. aegypti* from all localities were resistant to bifenthrin, lambda-cyhalothrin and permethrin with the exception of *Ae. aegypti* from Si Sa Ket in northeastern which demonstrated incipient resistance to permethrin. In addition, incipient resistance to alpha-cypermethrin was detected from all localities except for *Ae. aegypti* from Ubon Ratchatani in northeastern which was susceptible. Resistance to cypermethrin was only observed in *Ae. aegypti* from Phuket in southern, whereas susceptibility and incipient resistance of *Ae. aegypti* collected from other localities were observed. However, only *Ae. aegypti* from Si Sa Ket was susceptible to deltamethrin whereas *Ae. aegypti* collected from Ubon Ratchatani, Phuket and Krabi in southern demonstrated resistance. It was concluded that field-collected *Ae. aegypti* from all localities had developed resistance to the synthetic pyrethroids, especially bifenthrin, lambda-cyhalothrin and permethrin.

78 A comparison of attractants for *Stomoxys calcitrans* (Diptera: Muscidae) on dairy farms in Saraburi Province, Thailand

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Five attractant treatments (control, octenol, 10 kg of dry ice, mixture of cow dung and urine and mixture of 10 kg of dry ice and octenol) for collecting stable flies were compared on 2 dairy farms in Muak Lek district, Saraburi province. Five wk of trapping were carried out during the rainy season (from July to October 2013). Each trapping week was performed for 2 consecutive days. The daily trapping period was between 10 a.m. and 4 p.m. A total of 1,925 individuals of *Stomoxys* spp. were collected during July to October 2013 of which 1,058 were trapped on farm 1 and 867 on farm 2. Four species of *Stomoxys* were collected: *S. bengalensis* Picard, 1908, *S. calcitrans* (Linnaeus, 1758), *S. indicus* Picard, 1908 and *S. sitiens* Rondani, 1873. *Stomoxys calcitrans* was the most abundant species with 96% which was found on both farms. Dry ice was significantly more attractive for females of *S. calcitrans* than control, octenol, mixture of cow dung and urine. Both farms showed a difference between the numbers of male and female *S. calcitrans* collected by attractant types.

79 Indoor use of attractive toxic sugar bait (ATSB) to effectively control the malaria vectors, *Anopheles gambiae s.l.* in Mali, West Africa.

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Attractive toxic sugar bait (ATSB) methods are highly effective for outdoor mosquito control. ATSB solutions containing any gut toxins can be either sprayed on plants or used in simple bait stations to attract and kill sugar-feeding female and male mosquitoes. The studies were conducted in 5 villages located near the margins of the inland delta of the river Niger, Mali following the rainy season. To determine the rate of mosquito feeding on bait stations, 1 bait station per house containing attractive sugar bait (ASB) (without toxin) plus a food dye marker, was set up in 10 houses in each of the 5 villages. Pyrethrum spray collections (PSC) were then done after 1 day and the percentage of female and male mosquitoes that had fed was determined by visual inspection for the dye marker. Then, a 50-day field trial was done. In the experimental village, 1 bait station containing ATSB (1% boric acid active ingredient) was placed per bedroom (58 total bedrooms); no bait stations were placed in a comparable control village. Indoor densities of female and male *An. gambiae s.l.* were determined by PSC, and female mosquitoes were age graded. In the 5 villages, the percentages of *An. gambiae s.l.* feeding inside houses on the non-toxic bait stations containing the food dye marker ranged from 28.3 to 53.1% for females and 36.9 to 78.3% for males. Following ATSB indoor bait station presentation, there was a significant reduction in *An. gambiae s.l.* populations at the experimental village with a 90% reduction in female and 93% reduction in male populations. Moreover, the proportion of females that had undergone 4 or more gonotrophic cycles was reduced to 17.3% of the diminished population, compared to 33% at the control village.

80 Exploiting mosquito olfaction for the development of novel carbon dioxide mimic attractants

Samer Elkashef, selkashef@olfactorlabs.com, Michelle Brown and Molly Schmid

Mosquito-borne diseases continue to be a matter of life and death in the developing world where integrated pest management strategies are difficult to develop due to insufficient access to resources. In an increasingly interconnected world, mosquito-borne diseases traditionally thought of as foreign disease agents, such as dengue and more recently chikungunya, continue to emerge as public health concerns in the United States. An important front line defense in both cases is mosquito trapping, be it for identification of disease carrying insects or for abatement purposes. Recently, advances in the understanding of mosquito olfaction have shown that molecules other than carbon dioxide can activate the carbon dioxide receptor in mosquitoes providing alternatives to dry ice to be used in the field. Olfactor Laboratories Inc. (OLI) has been working on such technologies. Here we show that through the use of our proprietary screening algorithm and electrophysiology we are able to identify active formulations that activate the carbon dioxide receptor neuron of mosquitoes. Furthermore, we show that the efficacy of these lead formulations in trapping *Aedes aegypti* and *Culex quinquefasciatus* in semi-field conditions are nearly equivalent to carbon

dioxide with both the BG Sentinel and the CDC light trap. OLI's ultimate goal is the development of cost effective, easy-to-use attractants that can be used by both professionals and consumers.

81 Suitability of mixing fluorescent dye in adulticides and its impact on droplet characteristics and pesticide efficacy

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Fluorescent dyes are commonly used to help visualize insecticidal droplets or to trace movement of insecticides; however, the effect these dyes have on the insecticide's efficacy and droplet characteristics is unknown. This study evaluated the effects of mixing Uvitex OB fluorescent dye with 5 adulticides on their efficacy in a wind tunnel. Efficacy was determined via droplet size characteristics, spray flux (AI deposition), and female adult *Aedes aegypti* mortality. Fyfanon® ULV, Anvil® 10 + 10, Duet™, Aqualuer® 20-20, and Zenivex® E20, diluted with corn oil were tested with and without dye at maximum, minimum, and half minimum label rates. Adulticide droplet size was not affected by the addition of dye to any of the 5 pesticides. Mosquito mortality was strongly correlated with AI deposition for all pesticides except Duet. There was no difference among correlation coefficients of the 5 pesticides and between coefficients of any pesticide pairs indicating that all correlations were similar. The addition of dye non-significantly and non-consistently affected mortality. The source of this variability was due to large variation in mortality among different replicates of the same treatment.

Disclaimer: The views expressed in this presentation are those of the author and do not necessarily reflect the official policy or position of the Department of the Navy, Department of Defense, nor the U S Government.

82 Field evaluation of a new ULV mosquito adulticide against susceptible and pyrethroid tolerant *Culex* spp.

Gary Braness, gbraness@yespmc.com and Dennis Candito

A field trial evaluating a new ULV mosquito adulticide from Bayer CropScience was conducted in mid-July 2014 in Elk Grove, CA in cooperation with the Sacramento-Yolo Mosquito and Vector Control District. Applications of DeltaGard Public Health Insecticide (0.00089 lb AI deltamethrin/acre) and Anvil 10+10 ULV (0.0036 lb AI sumithrin/acre + 0.0036 lb piperonyl butoxide) were made over test plots using trucks mounted with London Fog model 18-20 gas powered foggers. Test plots consisted of 2 rows at 100, 200, and 300 ft downwind from the truck path. Each row was separated by 100 ft. Three mosquito cages were placed at each distance on stakes and in 1 row impingers with MgO or Teflon slides were placed adjacent the mosquito cages. Caged mosquitoes were set outside the treatment area to serve as controls. Each cage contained an average of 24 susceptible *Culex quinquefasciatus*, pyrethroid tolerant *Cx. pipiens*, or wild caught *Cx. tarsalis*. All mosquito cages and slides were collected at 30 min after application and returned to the laboratory. Knockdown efficacy was recorded at 1 h and mosquito mortality at 12 and 24 h after application. Percentage knockdown was greatest for the susceptible strain of *Cx. quinquefasciatus*. Mortality at 24 h for DeltaGard was 98 to 100% for all mosquito species at 100 and 200 ft. At 300 ft from the point of application, all *Cx. quinquefasciatus* were dead with 88% mortality for *Cx. pipiens* and *Cx. tarsalis*. Mortality at 24 h for Anvil 10+10 ULV ranged from 20 to 88% at 100 and 200 ft. At 300 ft, 64% mortality of *Cx. quinquefasciatus* was recorded, while mortality for *Cx. pipiens* and *Cx. tarsalis* was 3 and 8%, respectively. Results from impinger slides showed that spray droplets for both treatments reached the caged mosquitoes.

83 Transgenic mosquitoes: trials and tribulations

Derric Nimmo, derric.nimmo@oxitec.com, Andrew McKemey and Camilla Beech

Mosquito-borne diseases, such as dengue fever, chikungunya and malaria, are major and increasing international public health concerns. The two main vectors of dengue are *Aedes aegypti* and *Ae. albopictus* and current control measures are proving difficult against both these mosquitoes. In an Oxitec control program, transgenic male mosquitoes (male mosquitoes do not bite or transmit disease) are released continually over a wide area to mate with the target pest population; progeny from these matings die and the target population declines. Trial results over the past 4 yr will be presented to demonstrate how effective the technology has been at reducing *Ae. aegypti* around the world. Regulatory progress in the US towards a trial in collaboration with the Florida Keys Mosquito Control District (FKMCD) will be presented.

84 Efficacy of DeltaGard® on *Aedes* and *Culex* mosquito species in New Orleans, Louisiana

Cynthia Harrison, basecake@yahoo.com, Mieu Nguyen, Princeton King, Brendan Carter, Sarah Michaels, J.R. Rowland and Claudia Riegel

With the threat of emerging arboviruses like chikungunya, ULV insecticide applications are a key tool to prevent disease outbreaks and new products are necessary for effective mosquito control. In this trial, the efficacy of DeltaGard® was tested against 3 medically important species: *Aedes aegypti* (yellow fever mosquito), *Ae. albopictus* (Asian tiger mosquito), and *Culex quinquefasciatus* (southern house mosquito). All 3 species were field-collected as eggs during the summer of 2014 and reared at standard laboratory

conditions. Cages of 5-day old female mosquitoes were placed on stands at distances of 100, 200, and 300 ft from the site of application, in an open area without obstructions. DeltaGard® (Bayer CropScience LP, Austin, TX) was administered by truck-mounted ULV sprayer (ADAPCO Guardian 190 ES, Sanford, FL) first at a low-rate (4.5x10-4 lb deltamethrin/acre) and then mid-rate (4x10-4 lb deltamethrin/acre) application. Post low-rate application, mortality was highest amongst *Ae. aegypti* and *Ae. albopictus* while mortality of *Cx. quinquefasciatus* was varied (0-97%). Post mid-rate application, 100% mortality was observed in *Ae. aegypti* and *Ae. albopictus* and in *Cx. quinquefasciatus*, 100% mortality was achieved in all but 2 of the 9 locations. *Culex quinquefasciatus* populations in New Orleans have been previously shown to have a low level of resistance to pyrethroids and in this test DeltaGard® demonstrated significant treatment mortality amongst 3 common and medically important mosquito species.

Latin American Symposium I

85 ***Ae. aegypti* (L.) susceptibility to larvicides and adulticides in Puerto Rico**

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Aedes aegypti (L.) plays a critical role in dengue fever transmission. Dengue cases have recently been on the rise in Latin America, however the majority of control programs are faced with insecticide resistance. Susceptibility to insecticides of *Ae. aegypti* populations in Puerto Rico has not been characterized; the aim of this investigation was to evaluate larval susceptibility to Temephos® and spinosad (Natular® EC), and adults to permethrin. We also determined enzymatic detoxification levels for alpha and beta esterases, glutathione-s-transferases, oxidases, insensitive acetylcholinesterase and mutations associated with pyrethroid resistance. Our results show larval susceptibility to Temephos and spinosad, yet enzymatic levels were altered without correlation between LC_{50} and enzyme over expression ($p<0.05$). Adults presented resistance to permethrin not associated with enzymatic levels ($p<0.05$) and positively associated with both Cys 1534 and Ile 1016 mutations (frequencies between 0.8-1 and 0.65-1 respectively). These results highlight the need develop alternative strategies for vector control.

86 **Vertical distribution of sand flies (Diptera: Psychodidae: Phlebotominae) in Bacalar, Quintana Roo, Mexico**

Wilfredo Arque Chunga, wilfrach@gmail.com, Jorge Jesús Rodríguez Rojas, Ildefonso Fernández Salas and Eduardo Alfonso Rebollar Téllez

Sand flies of the genus *Lutzomyia* are small insects about 1.5 to 2.5 mm depending on the species, low flight capacity, there are different species capable of transmitting pathogens such as leishmaniasis, when feeding on reservoir hosts. In the world there are just over 700 known species; in Mexico, so far, a total of approximately 50 species of sand flies have been recorded. The immature stages develop in soils rich in organic matter. The adults are nocturnal with crepuscular activity; during the day, they remain in humid and dark environments, like tree cavities and caves. The presence of sand flies have been observed in the canopy of the trees with CDC light traps settled from 18:00 to 24:00 h in the Ejido Limones, Bacalar - Quintana Roo state, at 3 km from the nearest village. The CDC light traps were installed to 6, 8, 9, 10, 11, 12 and 15 m high, with a marked rope, for 3 consecutive nights during 4 mo. A total of 99 specimens were collected and preserved in 70% alcohol; the specimens were then mounted and identified. Of the total collections, 33% were males and 66% were females. The predominant species in all 7 different heights was *Lutzomyia cruciata*, which demonstrates the activity and/or presence of sand flies in the tree canopy in search of food sources for activity or development, these could be potential hosts of leishmaniasis.

87 **Resistance to pyrethroids in triatomine (Hemiptera: Reduviidae) vectors of the Chagas disease in Mexico**

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Chagas disease in Mexico is mainly transmitted by triatomine bugs, with 13 species implicated as vectors of *Trypanosoma cruzi*. In Mexico, there are no records of the status of susceptibility to insecticides used for their control, therefore the aim of this study was to determine the susceptibility of populations of *Triatoma pallidipennis*, *T. picturata*, *T. mazzottii* and *T. longipennis* to deltamethrin and permethrin. We used topical technique (WHO 2005) and determined LD_{50} and LD_{90}. Also, the biochemical mechanisms related with the resistance to pyrethroids such as: α and β esterases, glutathione-s-transferases (GSTs) and mixed function oxidases (MFO) .The presence of kdr type mutations was also determined (Fabro *et al*. 2013; Caprioti *et al*. 2014). Populations analyzed were resistant to deltamethrin and permethrin with RR $\geq$1000X by comparison with a susceptible strain (Piccolo *et al*. 2002). Furthermore, high enzymatic activity was implicated as well as the presence of 2 possible new kdr type mutations given by non-synonymous changes in the amino acids sequences.

88 Evolution of Ile1,016 and Cys1,534 replacements in the *para* gene in Mexican *Aedes aegypti* populations

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Massive use of pyrethroids has driven mosquito populations to develop insecticide resistance, threatening the success of vector control programs. Pyrethroid resistance in *Aedes aegypti* has been associated with replacements in the voltage-gated sodium channel (VGSC). One is located in codon 1016, with replacement of valine for isoleucine (I1016), and a second one is located in codon 1534, with a phenylalanine for cysteine replacement (C1534). In Mexico, I1016 allele frequencies increased from 2000 to 2008. We found that C1534 was also present in the same mosquito collections. In this study, we want to understand how these two mutations have segregated in the past. We performed a linkage disequilibrium analysis between I1016 and C1534. We also analyzed the haplotype composition in collections. We propose four hypotheses: 1) neither replacement affects fitness; the two replacements in VGSC arose separately and eventually became linked (*cis*) through recombination; 2) I1016 reduces fitness and requires C1534 in *cis* before it can become established, 3) C1534 reduces fitness and requires I1016 in *cis* before it can become established; or 4) both replacements affect fitness and require one another in *cis* to increase. Point mutations in codon 1016 and 1534 appear to have occurred independently. There was a rapid increase in the V1016/C1534 haplotype frequencies between 2003-2008 while I1016/F1534 frequencies barely increased. Besides, between 2003-2012, C1534 increased rapidly and became nearly fixed in populations whereas I1016 frequencies increased more slowly and only increased to a frequency of 0.729. We conclude that I1016 may confer lower fitness and require C1534 in *cis* before it can become established, supporting our second hypothesis.

89 Amino acids, acylcarnitines and organic acids profile and this correlation with insecticide resistance in *Culex quinquefasciatus* (Say)

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Several studies have been conducted on the biological fitness of resistant *Culex* spp. to synthetic and biological insecticides; however, no study has been carried out related to the metabolic cost associated with insecticide resistance. In this context, amino acids, acylcarnitines and organic acids involved in the completion of adult development and its effect enhancing the longevity in *Cx. quinquefasciatus* females has been studied. The aim of the present study was to investigate the fitness cost associated with amino acids, acylcarnitines and organic acids in resistant strains (surviving larvae to insecticides and not exposed) compared with a susceptible strain in order to understand the metabolic profile on resistant-mosquitoes. Using tandem mass spectrometry, concentrations of amino acids, acylcarnitines, and organic acids were differentiated in 2 experimental approaches: surviving larvae to a LC_{50} of permethrin, larvae without exposure to the insecticide and the susceptible strain not exposed to permethrin. The means, standard deviations, and ranges (upper and lower) were determined for the amino acids, acylcarnitines and organic acids of 5 field populations and the susceptible strain. Statistically significant differences were observed between all the experimental contrasts. The levels of short, medium and long chain acylcarnitines, amino acid and organic acid were also significantly different between groups. This is the first report establishing baseline levels of amino acids, acylcarnitines and organic acids and it could be considered as a powerful tool that sheds light on possible changes in genes involved in energy metabolism.

90 Semiochemical manifestations and enzymatic activity of *Salvia officinalis* on *Stegomyia aegypti* (Diptera: Culicidae)

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Essential oils (EOs) have demonstrated biological activity against the dengue vector *Stegomyia aegypti.* Activity larvicidal, pupicidal, repellency and inhibition of oviposition from *Salvia officinalis* EO on *Stg. aegypti* (Diptera: Culicidae) were evaluated. We used the Rockefeller colony of *Stg. aegypti* for laboratory bioassays. Larvae between 3rd and 4th instars were subjected to different concentrations that cause mortality at 24 h and the other at 48 h. Insecticidal activity against larvae was assessed at exploratory concentrations (EC) of 250, 310 and 390 ppm, pupicidal activity at 250, 310 and 390 ppm. Adulticidal activity was estimated with CDC method of 30, 300, and 1,000 ppm. The repellent activity was determined with the number of mosquito bites and landings. The deterrent activity of oviposition was estimated at EC 5, 50 and 200 ppm. Enzymatic tests *in vitro* were performed to determine the mechanism of metabolic action. *Salvia officinalis* showed greater insecticidal activity against larvae at 24 and 48 h at the 63 ppm and 76 ppm (33.7±17.6% and 47.5±24.04%); the pupicidal activity was registered at 310 ppm and 390 ppm at 48 h (88,89±1,53% and 100±0%); and the adulticidal activity effect was detected at 300 ppm with 57.5±0%. The repellent activity was 53.85±1%. The deterrent activity of oviposition at 200 ppm was 97±4.81%, with oviposition

activity indices (OAI) of -0.94, showed negative oviposition for the females. *In vitro* enzymatic tests on larvae treated at 45, 55 and 80 ppm, showed an activity decreasing for enzyme acetylcholinesterase (AChE). The EO form *S. officinalis* provides insecticidal activity and affects the enzymatic metabolism of *Stg. aegypti*.

91 Adulticide lethal traps: a potential system for residual effect of indoor populations of *Aedes aegypti*, laboratory evaluation in Tapachula, Chiapas, Mexico

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Insecticide spraying is the most common and representative vector control method of *Aedes aegypti* adults. Among the variety of methods, most lack long-lasting effects. This project evaluates lethal traps with residual effect as a complementary method of ULV application in residential areas. The trap is a black cardboard box with cut out slots for mosquito entry, baited with BG-Lure™ (Bioagents) artificial human sweat. We evaluated 2 different control methods: First, Netprotect®, a deltamethrin-impregnated net and second, a removable sticky sheet. Capture – release tests were performed in a cage of 2 m^3 with 100, 3-6 day old females and an exposure time of 24 h. Captured mosquitoes and mortality were accounted. Five replicates were performed for each trap using a new batch of mosquitoes for each repetition. There was no significant difference in mortality between both traps ($t=-0.68$, $p > 0.05$). This novel approach has the potential to increase the effectiveness of current vector control methods.

92 Insecticide resistance monitoring in *Aedes aegypti* from Mexico through CDC bottle bioassay

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Pyrethroids have been widely used in Mexico to control *Aedes aegypti*, the main vector of dengue viruses in different parts of the country. Control activity represents an important way to select insecticide resistant populations if the susceptibility status of the populations is unknown. In this context, the Centers for Disease Control and Prevention (CDC) bottle bioassay is a surveillance tool for detecting resistance to insecticides in vector populations. This technique is designed to determine if an insecticide and/or a particular formulation is able to control vector populations at a specific location at a given time. Bottle bioassay was used to detect insecticide resistance in *Ae. aegypti* populations from Mexico to 5 different insecticides (3 pyrethroids, 1 carbamate and 1 organophosphate). Also 1 set of mosquitoes, which survived pyrethroid exposure, were used in molecular assays to detect kdr mutations. The results of bioassays, combined with molecular assays, can assist in determining which insecticide could be used in populations studied and if rotations or mosaics of insecticides are viable for the sustainable use of insecticides in control programs of *Ae. aegypti* at the local and/or regional level.

93 How to: aerial mosquito field trials

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For most county and parish-wide area abatement programs in the US, aerial mosquito spraying is the most efficient space spray, covering large acreage areas faster than by ground ULV alone. Unfortunately, aerial spraying effectiveness has been reduced because of product dispersion through offset, and above all aircraft conditioning. If not optimized, field trial work can be significantly compromised. This presentation will discuss optimization of aerial spraying, explaining the essentials of a successful aerial trial, starting with area selection, how to get your product to the intended target, techniques and equipment, spraying systems improvements, cage locations, and inclinations and droplet collectors as indicators of spray density and volume median diameter (VMD). Historical case scenarios will illustrate the major technical advancements.

93a Comparison of traps for catching Phlebotominae (Diptera: Psychodidae) in an endemic focus of leishmaniasis in southern Mexico

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Some species of phlebotomine sand flies are important vectors of pathogens such as *Leishmania* sp., *Bartonella bacilliformis* and several arboviruses in America. Therefore, it is important to assess and know trap differences for the efficient capture of these insects. In this 12 mo study, 11 different traps (CDC-I: CDC incandescent light trap, CDC-B: CDC white LED (light-emitting diode), CDC-A: CDC blue LED, CDC-V: CDC green LED, CDC-R: CDC red LED, Disney trap, Disney-B: Disney white LED, Adhesive trap, Adhesive-B: Adhesive white LED, Delta trap, Delta-B: Delta white LED) were run for 3 consecutive nights each month from 1800-2400 h, using a randomized block design, in a town in southern Mexico. A total of 1,020 specimens were collected (82.16% females and 17.84% males) of 8 species (*Brumptomyia mesai*, *Lutzomyia carpenteri*, *Lu. cruciata*, *Lu. deleoni*, *Lu. longipalpis*, *Lu. olmeca olmeca*, *Lu. ovallesi* and *Lu. shannoni*). Of the total, average monthly captures in descending order were: CDC-I (23.75 ± 4.35), CDC-B (15.08 ± 3.18), CDC-V (12.92 ± 2.23), CDC-R (12.42 ± 2.41), CDC-A (12.42 ± 3.49), Disney (5.50 ± 2.39),

Disney-B (1.17 ± 0.37), Delta-B (1.17 ± 0.60), Adhesive (0.17 ± 0.11), Adhesive-B (0.17 ± 0.11) and Delta (0.08 ± 0.08). The 5 CDC light traps showed greater catch compared to the other traps and within the CDC, the incandescent light trap captured more. The attraction for each species by the different traps is discussed.

Looking In Our Rear View Mirror: Events, Personalities and Discoveries That Have Shaped Mosquito Control II

94 H, D, and K: Who were those guys?
Tom Wilmot, wilmotg@charter.net

Leland Ossian Howard, Harrison Gray Dyar and Frederick Knab provided significant contributions to our early understanding of the biology, control and taxonomy of new world mosquitoes. L.O. Howard served many years as Chief Entomologist at the US Department of Agriculture and Consulting Entomologist with the USPHS and was the first man elected as Honorary Member of AMCA. Harrison Dyar spent many years as custodian of the US National Museum's collection of Lepidoptera and in the USDA Bureau of Entomology. He served as editor of the *Journal of the New York Entomological Society*, one of the most prestigious journals of his day, and started his own journal *Insecutor Inscitiae Menstruus*. Frederick Knab was a widely respected artist and field biologist. These men collaborated to produce the 4-volume treatise "The Mosquitoes of North and Central America and the West Indies". Together, they described over 600 mosquito species. This presentation provides a brief review of the lives of these men and how they fortuitously came together at the dawn of medical entomology.

95 A historical perspective of our understanding of the relationship between mosquitoes and bats
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With the exception of a few species found in the Southwest US that feed on nectar, pollen and fruit, the 40 bat species of the United States are exclusively insectivorous. Depending on the species, bats are predators of insects ranging in size from small soft-bodied flies, midges, mosquitoes and mayflies, to relatively large beetles and moths. Because mankind has witnessed bats foraging in the evening sky for thousands of years and the eventual verification of mosquitoes in bat stomachs, generalizations eventually emerged crediting bats as natural control agents of mosquitoes and their subsequent benefit to public health. To this end, 2 attempts were made in the early 1900s to construct massive-sized bat houses in Texas and Florida with the premise that thousands of local bats should control millions of local mosquitoes thereby reducing the threat of encephalitis and malaria. But the relationship between bats and mosquitoes is best described as unpredictable and varying widely. Few studies exist that attempt to empirically define bat-mosquito interactions and these studies measure only specific locales within the specific environmental conditions of those areas. Presently, a casual surf on the internet on this topic produces a mostly non-scientific offering extolling the benefits of *natural* mosquito control via local bat colonies and the employment of bat houses towards this end. Perhaps the non-scientific assumption that bats help control mosquitoes is a case of the "end justifying the means". Certainly, bats need all the protection we can afford them, especially in light of the recent catastrophic result of white nose syndrome. Nevertheless, in the interest of sound public health and professional vector management programs, effective insect repellents and protective clothing should not be discontinued, even if the most well-occupied bat house -- or even a large cave -- are present in a neighborhood.

96 Willard V. King: mosquito warrior extraordinaire
Gordon Patterson, patterso@fit.edu

Willard van Orsdel King's (1888-1970) work as medical entomologist contributed greatly to understanding insect-borne diseases and the advance of mosquito control in the twentieth century. While still an undergraduate at Montana State University, King launched his career as a medical entomologist by isolating the role of ticks in the transmission of Rocky Mountain spotted fever. After earning his Ph.D. at Tulane University in 1915, King served the United States Department of Agriculture (USDA) laboratory in Mound, Louisiana as a researcher where he pioneered use of airplanes in the distribution of pesticides in the 1920s and taxonomy of mosquitoes of the Southeast. Later, King directed the USDA Orlando Parramore Street Laboratory that would become famous for its role in the development of DDT and the discovery of DEET. During World War II, King served as one of the leaders of the US Forces in the Southwest Pacific campaign against malaria and other mosquito-borne diseases. King's career as a mosquito warrior exemplifies a commitment to science and the public's welfare.

Student Competition III

97 Bacterial identification from local mosquitoes found in south Texas (*Aedes aegypti* and *Aedes albopictus*) and confirmation of uptake during larval stage

Fabiola Cantu, fcantu1@broncs.utpa.edu, Kristine Lowe, Erin Schuenzel and Christopher Vitek

Insects host a variety of microorganisms that colonize their tissues. Occurring mainly in the digestive tract, they probably exceed the number of cells in the insects themselves. These microorganisms live and multiply in the insect, contributing to digestion, nutrition and development of their host. Questions about bacteria within the mosquito midgut remain largely unanswered. Microbes in the aquatic habitat serve as a food source for larvae; however, the question arises whether these microorganisms are being up taken during the larval stage. Previous research suggests microorganisms are ingested during the larval stage of the mosquito life cycle. This has yet to be proven, which indicates the importance of confirming whether larvae ingest microbes from their particular environments. Previous works have reported isolation of microorganisms from the midgut of *Culex quinquefasciatus*, *Anopheles dirus* and *Anopheles albimanus*. We observed differences in midgut microbial communities inhabiting local mosquitoes found in south Texas. Data were obtained using16s rDNA methodology following the dissection of individual midguts. Results regarding the bacterial uptake during the larval stage are discussed based on the use of GFP expressing bacterial strains with fluorescent microscopy. In the context of mosquito control, this research sheds light on the role bacterial communities have on the vectors of many diseases.

98 Genetic basis of insensitivity to DEET in *Anopheles gambiae*

James Ricci, jricc001@ucr.edu and Bradley White

Despite widespread and prolonged use, insensitivity to DEET has rarely been observed in mosquitoes, and DEET's mode of action upon mosquitoes is still debated. We demonstrate significant variation in DEET insensitivity in a population of *An. gambiae*, and that this heritable variation can be selected for over multiple generations to produce replicate divergent lines. Genomic analysis of selected lines to reveal genes responsible for DEET insensitivity, and its mode of action, is discussed.

99 A spatially-explicit, rule-based approach to predicting and managing mosquito populations

Daniel Dawson, dan.dawson@ttu.edu and Christopher Salice

We demonstrate a novel, spatially explicit, and flexible approach to making mosquito control decisions using fuzzy logic rules in a Geographic Information System environment. In an example, we generate potential mosquito density surfaces by considering habitat associations, dispersal patterns, and environmental influences on mosquito ecology. By incorporating human density information, we create a simple risk index that shows where mosquito control treatments may be applied for best affect, and how risk surfaces change when treatments are made.

100 Evaluation of surveillance methods for *Aedes* vectors of La Crosse virus in eastern Tennessee, USA

Cassandra Urquhart, curquhar@vols.utk.edu, David Paulsen, Abelardo Moncayo and Rebecca Trout Fryxell

La Crosse virus (LACv) is primarily transmitted via the bite of infected *Aedes triseriatus,* and may be transmitted by bites from *Ae. albopictus* and *Ae. japonicus.* Recent La Crosse encephalitis case clusters in Tennessee prompted investigations in Appalachia. This study compared the ability of five adult traps to collect LACv vectors and determined the prevalence of LACv in Knox County Tennessee mosquitoes. At eight urban sites, several adult traps operated; a BG-sentinel trap, two CDC traps (baited with dry ice or dry ice and chemical lure), a gravid trap, and a resting trap. Two oviposition traps per site operated for additional surveillance. Mosquitoes were identified, pooled, and sent to the Tennessee Department of Health for LACv screening via RT-PCR. A total of 3,989 adult LACv vectors were collected; 3,486 *Ae. albopictus* (619 pools), 398 *Ae. triseriatus* (136 pools), and 99 *Ae. japonicus* (61 pools). The most effective collection method for *Ae. albopictus* was a CDC trap baited with dry ice and lure, for *Ae. triseriatus* it was a BG trap baited with CO_2, and for *Ae. japonicus* it was a gravid trap. Eight adult pools tested RT-PCR positive for LACv (seven were *Ae. triseriatus* and one was *Ae. albopictus*). All *Ae. japonicus* pools tested LACv negative. Oviposition traps yielded 951 mosquitoes reared from egg papers and all were LACv vectors (589 *Ae. albopictus*, 355 *Ae. triseriatus*, 7 *Ae. japonicus*) and all tested negative for LACv. Results from this study indicate that multiple traps should be used when monitoring LACv vectors to ensure accurate vector and virus prevalence data. This information enhances the knowledge base of LACv and its vectors in eastern Tennessee, and with adoption and implementation will improve control measures and disease prevention.

101 Determining suburban adult mosquito diversity through various sampling techniques

Christopher Holderman, chrish2@ufl.edu, Salvador Gezan, C. Roxanne Connelly and Phillip Kaufman

To help elucidate when and which species of mosquito is transmitting the causative agent of dog heartworm disease (*Dirofilaria immitis*), a sampling project was established to determine phenology and diversity of

mosquitoes in suburban Gainesville, FL. Weekly, 3 active techniques (sweep netting, and two styles of aspirators) were used in conjunction with a CO_2 baited miniature CDC light trap to sample for adult mosquitoes. One year of abundance data and possible vectors will be presented.

102 The oviposition of *Aedes albopictus* in response to Copepoda in field conditions
Jimmie Teague, jlteague@uncg.edu and Dr. Gideon Wasserberg

Biological control methods can play an important role in the prevention of vector-borne diseases. To be effective, the control agent should act as bait or not be repellant. The copepod *Mesocyclops longisetus* has been used as a control agent for mosquito larvae. However, the ability of female mosquitoes to detect and respond to its presence in terms of oviposition site selection is poorly understood. In this study, we used *Aedes albopictus* and assessed ovipositional site response and its effect on hatching rates in the presence of the predator, Copepoda. We hypothesized that the presence of the predator will have a negative effect on the oviposition, as well as on the hatching rates. If the location for possible oviposition is the habitat of a natural predator, then the mosquito will show preference for another location. We conducted this experiment by establishing four, 100-meter transects, each containing a set of mosquito egg collecting cups, in Peabody Park in Greensboro, NC. These oviposition traps were randomly placed in pairs of treatment and control. The treatment cups contained various densities of Copepoda, either 50 or 100, and the control cups contained distilled water. The cups were deployed in 5-day intervals, then collected and analyzed. The results showed that there was not a significant difference in oviposition between cups. Nor was there an effect on the hatching rates in the presence of a predator. These results showed promise with the gravid females not responding to a foreign enemy. This illustrated a positive relationship in population control of nuisance agents by the use of natural predators. By passively controlling and reducing the population of vector species, the risk of several mosquito-borne diseases can be mitigated.

103 Pyriproxyfen for mosquito control: female sterilization or horizontal transfer to oviposition substrates by *Anopheles gambiae* sensu stricto and *Culex quinquefasciatus*
Oscar Mbare, oscarmbare@gmail.com, Steve Lindsay and Ulrike Fillinger

Pyriproxyfen was suggested for the control of disease vectors as a sterilizing agent and for auto-dissemination to aquatic habitats. We explored if and at which time of exposure (i.e., before or after a blood-meal) *Anopheles* and *Culex* females became sterilized and whether this affected their ability to transfer the insecticide. Exposure 24 hours around blood-feeding reduced egg-laying by 84%. Sterile females did not visit the egg-cup. For successful auto-dissemination the optimum time for contamination is close to oviposition.

Adult Control III

104 Evaluation of Duet® and Mosquito Master® 412, mosquito adulticides with distinct modes of action against field collected mosquitoes in Maricopa County, Arizona
Jacob Hartle, jhartle@clarke.com and Kirk Smith

Understanding how products with distinctly different modes of action can affect a district's wild adult mosquito population is a key principle of an integrated pest management (IPM) program. Trials conducted in Maricopa County, AZ, evaluated Mosquito Master® 412, an organophosphate formulation containing 12% chlorpyrifos and 4% permethrin; and Duet®, a pyrethroid formulation containing 1% prallethrin, 5% sumithrin, and 5% piperonyl butoxide. Evaluations consisted of formulated bottle bioassays and ground ultra-low volume caged mosquito field trials for both formulations against wild-caught mosquitoes containing, *Culex quinquefasciatus*, *Cx. tarsalis*, *Aedes vexans*, and *Psorophora columbiae*. Results of the bottle bioassay with a formulated sumithrin product showed 100% mortality at 40 and 45 min for 2 replicates. Results with chlorpyrifos showed 100% mortality at 80 and 50 min for 2 replicates. The adulticide Duet was applied at 0.76 fl oz/acre, the results of 24 h mortality was 84, 88, and 89% for 3 replications. The results of the adulticide Mosquito Master 412 showed 24 h mortality of 100, 100, and 84% for 3 replicates. These results showed that both products are effective adulticide options for this district. Environmental conditions during both ground trials will be discussed.

105 Resistance characteristics of *Anopheles gambiae* Akron strain mosquitoes and resistance-breaking trifluoromethylphenyloxime carbamates
Jeffrey Bloomquist, jbquist@epi.ufl.edu, Troy Anderson, Derek Craft, James Mutunga, Daniel Swale, Fan Tong, Dawn Wong and Paul Carlier

Anopheles gambiae is one of the major vectors of malaria in Africa. Insecticide use for vector control has selected for resistance, and the Akron strain that carries multiple resistance mechanisms has been isolated from Benin. We report resistance ratios for permethrin, deltamethrin, and DDT on this strain, as well as a range of carbamates. In addition, biochemical characterization of metabolic resistance arising from cytochrome P450 monooxygenases, carboxylesterases, and glutathione-*S*-transferases found that all were

elevated in the Akron strain. Substituted acetophenone oxime carbamates were toxic to both susceptible and resistant Akron mosquitoes, having resistance ratios of 5 or less, which was much less than the resistance observed for standard commercial carbamates. These compounds are valuable leads for new carbamate insecticides that are effective in controlling both susceptible and resistant mosquito populations.

106 Ground mosquito field trials: What is your problem?
Grifith Lizarraga, glizarraga@clarke.com, Derek Drews and Jake Hartle

Mosquito field trials have been the backbone for efficacy and resistance testing, equipment, and techniques. The intent of this presentation is to explain the basic procedures to effectively accomplish ground field trials including techniques and equipment used, discussion on specific improvements in mosquito cages, weather-friendly support equipment (e.g., wind-driven mosquito cage holders), compact weather station collectors, and droplet reading techniques. Historical case scenarios will illustrate the major technical advancements.

107 Silencing trehalose-6-phosphate synthase incapacitates adult mosquitoes by interfering with the biosynthetic pathway for flight fuel
Alden Estep, Alden.Estep@ARS.USDA.GOV, Carolyn Talcott and Julia Moore

Trehalose is a disaccharide comprised of 2 glucose molecules. It is the main blood sugar of insects and is essential for flight. Trehalose is synthesized by 2 enzymes: trehalose-6-phosphate synthase (T6PS) converts glucose-6-phosphate to trehalose-6-phosphate, and trehalose-6-phosphate phosphatase (T6PP) dephosphorylates trehalose-6-phosphate to form trehalose. Because trehalose is the principal carbon source for flight energy and is absent in mammals, the biosynthetic pathway for trehalose is an attractive target for novel insecticide chemistry. We found that microinjection of small interfering RNA (siRNA) of T6PS – but not T6PP – rendered 80-100% of female *Aedes aegypti* incapable of righting themselves from an inverted position for 6 h and induced flightlessness in >60% for up to 12 h. Ablation of T6PS resulted in 30% mortality after 24 h. We will discuss the potential control benefit of flightlessness even in the absence of mortality. These results support T6PS inhibition as a rational mosquito control strategy.

108 Aerial ULV application of Dibrom against *Aedes aegypti* in simulated urban and rural residences
Seth C. Britch, seth.britch@ars.usda.gov, Kenneth J. Linthicum, Robert L. Aldridge, Mark S. Breidenbaugh, Mark Latham, Peter Connelly and Jerry D. Kerce

Reaching endophilic *Aedes aegypti* mosquito vectors of dengue located in human residences with aerial ULV pesticide applications is a prominent complication in operational wide area public health mosquito control activities. We conducted trials with a military C-130 fitted with a modular aerial spray system and a Hughes MD 500D helicopter fitted with a high pressure spray system to apply Dibrom (naled) over simulated urban and rural residences on a military reservation in Florida against caged adult sentinel colony-reared *Ae. aegypti* mosquitoes. We placed sentinel mosquitoes in open and protected locations both inside and outside of residences and measured mortality following multi-swath aerial applications conducted with typical real-world operational parameters. However, meteorological conditions were cool to warm with moderate humidity, and not representative of the hot, high humidity typical of dengue endemic regions. Selected residences were partially sealed to provide an additional challenge to the ULV spray. Dibrom from the C-130 application penetrated well-ventilated residences and caused limited mortality in sentinel mosquitoes inside several protected locations; mortality in protected and unprotected outdoor locations was substantially higher, and approached 100% in unprotected outdoor locations. Dibrom from the MD 500D application penetrated well-ventilated residences and caused high mortality in many unprotected indoor and outdoor locations, but caused limited mortality in 1 indoor and 2 outdoor protected locations. Potential effects on spray dispersion of different droplet size spectra from the 2 spray systems and of different air currents generated by the aircraft are discussed.

109 Development of a multi-rotor unmanned aerial system for mosquito control operations - Part I
Gregory Williams, gwilliams@hudsonregionalhealth.org, Randy Gaugler, Scott Crans, Rafael Valentin, Devi Suman and Yi Wang

Unmanned aerial vehicles (UAVs) are currently being used for everything from package delivery to search and rescue operations. However, experts predict 80% of the commercial UAV market will come from precision agriculture, generating over 100 billion dollars over the next 10 yr. This includes precision mosquito control. Our team has been exploring the use of UAVs for mosquito control and has developed a small inexpensive hexacopter for autonomous mosquito control missions. This talk will focus on our use of UAVs for mosquito larvicide missions. We will discuss recent improvements to the spray system and present flow rates, swath width and droplet data. Several autonomous operational trials were conducted over salt marsh habitat with VectoBac 12AS and we weigh the various larviciding strategies such as blanket treatments versus GPS mapped pool-to-pool applications. We will also recount the accuracy of the flight plans and efficacy of the applications.

110 Development of a multi-rotor unmanned aerial system for mosquito control operations - Part II
Randy Gaugler, gaugler@rutgers.edu, Gregory Williams, Scott Crans, Rafael Valentin and Yi Wang

Unmanned aerial systems (UAS) may offer enormous operational potential, but only if specific tasks can be successfully delegated to them. In Part I, we described a heavy-lift hexacopter as our aerial platform for a larvicidal sprayer module. Here we describe early efforts to design additional quick disconnect modules carried by our multi-rotor aircraft for mosquito surveillance and control activities. An ULV sprayer module for adulticides has been constructed and tested in a 3 X 3 field cage experiment. A double net module for adult collection at different altitudes has been flown successfully. A rotating 'bottle' sampler module for collecting larvae has been fabricated and lab tested. And a waterproof quadcopter has been constructed to transport a waterproof camera which permits image capture and possibly automated counting of immatures. These modules await proof-of-concept field validation. Pros and cons of multi-rotor UAS for mosquito operations are outlined.

111 How to: aerial mosquito field trials
Grifith Lizarraga, glizarraga@clarke.com, Derek Drews and Jake Hartle

For most county and parish-wide area abatement programs in the US, aerial mosquito spraying is the most efficient space spray, covering large acreage areas faster than by ground ULV alone. Unfortunately, aerial spraying effectiveness has reduced product dispersion through offset, and above all aircraft conditioning. If not optimized, field trial work can be significantly compromised. This presentation will discuss optimization of aerial spraying, explaining the essentials of a successful aerial trial, starting with area selection, how to get your product to the intended target, techniques and equipment, spraying systems improvements, cage locations, and inclinations and droplet collectors as indicators of spray density and volume median diameter (VMD). Historical case scenarios will illustrate the major technical advancements.

112 Effect of nozzle discharge direction on efficacy of ULV space spray
Alice Fulcher, aliamcd@bellsouth.net, Jennifer Wright, Michael Smith, Jeremy Anderson and Rui-De Xue

Field study for the effect of nozzle orientation of a truck mounted ULV sprayer on application efficacy through mortality of *Aedes aegypti* was conducted at St. Johns County Fairground, Elkton, FL during summer 2014. Two rows of bio-assay cages were set at 0, 50, 100, 200, 300, and 400 ft from the spray line. Cages were placed 5 ft off the ground in order to cover the area in which mosquitoes would generally fly. Control cages were deployed well out of the spray zone. Aqualuer 20-20 was applied at the maximum label rate with the ULV sprayer driving at 10 mph. Treatments included spray delivery with the nozzle pointed 45° upward, horizontal (0°) and 30° downward. Applications for 1 of the 3 replications with each nozzle orientation were made in a day. Knockdown after 10 min and mortality after 24 h was recorded.

The knockdown was not significantly different at any angle except from 45° at 0 ft distance, which was lower than from other angles. The mortality results showed complete mortality from horizontal nozzle up to 400 ft except no mortality for 1 location at 400 ft in 1 replication. The mortality at 30° angle was non-significantly lower than horizontal at 0, 200, 300, and 400 ft. The mortality from 45° angle was significantly less than that from other angles at 0 ft. The same angle had significantly less mortality (20.2 – 28.4%) than horizontal (83.3 – 100%) at 300 and 400 ft. This mortality, however, was not significantly less than that from 30° downward angle. Overall, horizontal nozzle showed best efficacy followed by 30° downward angle and then 45° upward.

Disclaimer: The views expressed in this presentation are those of the author and do not necessarily reflect the official policy or position of the Department of the Navy, DOD, nor the US Government.

Latin American Symposium II

113 Temephos and permethrin resistance in *Aedes aegypti* from Tapachula, Chiapas, México
Arturo Acero Sandoval, navio81@hotmail.com, Karla Saavedra Rodríguez, Américo Rodríguez Ramírez and Bill Black IV

Aedes aegypti mosquito is the main vector of dengue in the Americas. Dengue outbreaks are controlled primarily by elimination/reduction of mosquitoes and immature stages and chemical control (insecticides). In the last decade, pyrethroid insecticides have been strongly used, leading to high levels of resistance. In this study we used larval and bottle bioassay to assess the levels of temephos and pyrethroid resistance in 3 *Ae. aegypti* collections from Tapachula, Chiapas, Mexico in 2014. The lethal concentration that kill 50% of the mosquitoes (LC_{50}) in each locality was calculated, using the New Orleans strain as reference. We also determine the allele frequency for 2 mutations (Ile1016 and Cys1534) associated with knockdown resistance (kdr) in the voltage-gated sodium channel gene. Temephos LC_{50} was 0.87, 1.03 and 1.10 µg/ml for Centro Nte/Ote, Feb. 5 and Parlange, respectively. The LC_{50} obtained for permethrin ranged from 9.94, 11.19 and 4.49 µg/bottle in Centro Nte/Ote, Feb.5 and Parlange, respectively. New Orleans reference LC_{50} was 0.016

µg/ml and 0.6 µg/bottle. The allele frequencies for Ile1016 were 0.52, 0.62 and 0.51 for Centro Nte/Ote, Feb. 5 and Parlange, respectively. Higher allele frequencies were calculated for Cys1534 ranging from 0.89 to 0.98. Comparing the frequency of Ile 1016 with the response to permethrin in adults, we found that the Parlange population has the lowest gene frequency (0.89) and is more susceptible to permethrin (4.49 ul/bottle) than populations of CentroNte / Ote (9.94) and Feb. 5 (11.19).

114 Entomological survey for chikungunya virus on *Aedes aegypti* and *Aedes albopictus* mosquitoes in border communities from Suchiate, Chiapas, southern Mexico

Esteban Eduardo Diaz Gonzalez, dige90@gmail.com, Iliana Rosalia Malo Garcia, Alicia Dorantes Delgado, Maricela Laguna Aguilar, Tomas Nepomuceno Mejia, Rocio Ramirez Jimenez, Rosa Maria Sanchez Casas and Ildefonso Fernandez Salas

Chikungunya fever (CHIKV) is the new emerging arboviral disease in America transmitted by *Aedes aegypti* and *Ae. albopictus.* This virus of the family Togaviridae, genus *Alphavirus*, though it is not lethal, produces polyarthralgias that may incapacitate patients for weeks. Currently there is an outbreak in El Salvador and Guatemala, with 16,813 probable and 103 confirmed cases combined. In Mexico, there is a strong concern about the southern border with Guatemala due to uncontrolled migration from these countries. Additionally, Mexican surveillance system is focused on patients with less than 8 days of febrile outset in order to detect CHIKV by RT-qPCR and most of them may be discarded for testing. The reason for this is because most of clinics and hospitals lack of ultrafreezers for storing serums, especially in poorest states like Chiapas. The aim of this study is to carry out entomovirological surveillance for CHIKV in the southern border of Chiapas, Mexico. The advantage of this approach is that mosquitoes remain infected the rest of their lives, increasing the probability of detecting and isolating the virus. The sampling started in early October, and at this moment, 1,255 *Ae. aegypti* mosquitoes have been collected in 86 households of Suchiate localities. The surveillance is going to continue the rest of this year and we will have results before this work is presented.

115 Parity rates of *Anopheles (Kerteszia) neivai* (Diptera: Culicidae) and their importance in the transmission of malaria in the Pacific Colombian mangroves

Deiby Rodriguez, nefila27@hotmail.com, Jesús Escobar, Martha Quiñones and Ranulfo Gonzalez

Malaria is considered one of the most important diseases in Colombia, due to the geographical conditions that favor the development of vector species. Among them *Anopheles* (*K*) *neivai* has gained importance due to its anthropophilic behavior and ability to transmit the malaria parasite in wild environments. The aim of this research, realized between 2009 to 2011, in a municipality of the Pacific coast of Colombia, was to evaluate the physiological age of mosquitoes of *An. neivai*, and their possible influence on the transmission of malaria. A total of 42 sampling sessions during 4 expeditions were made. By human bait collections, abundance and time biting activity was recorded at each site sampling. Of a total of 4,256 specimens, 2,873 were collected in mangrove, 1,253 in canoes and 130 in the peridomicile. The peak of activity of *An. neivai* was observed between 18:00 and 19:00; however, this species showed activity for 24 h in the mangroves. The dissection of 570 females collected during the peak of biting activity, to determine the percentage of parous (multiparous) in the mosquito population, showed that 42.5% of the females had realized at least 1 oviposition. The correlation analysis showed there is no correlation between the density and the parity rate, which indicates that the number of parous females in the population is not dependent of the amount of mosquitoes dissected, and therefore the results obtained provide an approximate physiological age of the population. These results indicate that the females may actually be a potential risk for the transmission of malaria among the inhabitants of the area, especially in times during their performed work activities in the estuaries and mangroves.

116 Variation in putative cellular receptors of flavivirus in *Aedes aegypti*

Miguel Moreno-Garcia, Miguel.Moreno_Garcia@colostate.edu and William Black IV

During invasion of mosquito tissues and cells, the dengue virus (DENV) must attach to the midgut epithelial cells via one or more cellular receptors. It has been commonly assumed that the entry of DENV into cells is a well-conserved mechanism; nonetheless, populations of mosquitoes differ in their susceptibility to DENV infection. This variability could be associated with variation in midgut cellular receptors. Currently, by using multiple approaches and different cell lines, numerous candidates for dengue virus receptors have been provided. However, the genetic variation of this putative receptor has not been analyzed. The objective of this work is to characterize sequence variation in 8 different classes of receptors of *Ae. aegypti aegypti* (*Aaa*) and *Ae. aegypti formosus* (*Aaf*). For this, we compared variation, the numbers of nonsynonymous and synonymous substitutions (dN/dS ratio) and FST values per receptor class within and among 4 different populations (for *Aaa*: Merida-Mexico; Pai Lom-Thailand; for *Aaf*: Pk10 forest-Sengal; Koalack-Senegal). The dN/dS ratio is expected to be greater than 1 only if natural selection promotes changes in the protein sequence; meanwhile a ratio less than 1 is expected only if natural selection suppresses changes in the receptors through purifying selection. The differences among sequences could represent polymorphisms segregating in a single population. It is important to understand the relationship between selection pressures and biological variation in natural mosquito populations.

117 Spatiotemporal analysis of the comparison between dengue fever cases and vector detection by ovitraps and index mapping in Medellin, Colombia

Marcela Quimbayo, marceladelpilar45@gmail.com, Guillermo Rua, Wiliam Sanabria, Jose Chauca and Enrique Henao

In Colombia, dengue fever is an important disease in terms of public health, principally owing to the large number of cases that occur each year and the fact that they are distributed throughout the country's major urban centers. Currently, *Aedes aegypti* as well as *Ae. albopictus* are found in the city of Medellin, but only the former has been implicated as a vector of dengue fever. Health authorities in Medellin developed vigilance programs for the vector by mapping entomological indices and monitoring *Ae. aegypti* using ovitraps. However, to generate adequate vigilance strategies for prevention and control of the disease, it is necessary to determine the spatiotemporal relationship between the occurrence of dengue fever and the density of the vector. Therefore, the present study was carried out with the objective of evaluating the relationship between cases of the disease and the presence of the vector utilizing 2 sampling methods: mapping of conventional entomological indices and observation of the vector with ovitraps. Epidemiological and entomological data were applied for 2012 - 2014, and a spatiotemporal analysis between the different variables was performed. It was observed that the presence of both *Ae. aegypti* and the occurrence of the disease showed a highly heterogeneous pattern. However, the exact correlation between the epidemiological and entomological variables was not determined for all of the neighborhoods in the city. The results obtained from the study make it possible to provide entomological information that facilitates opportune decision-making by health authorities for the control of the disease in Medellin city.

118 Association of genes Dicer-2 and Argonaute-2 with vector competence of DENV-2 among *Aedes aegypti* (L.) populations of Veracruz, Mexico

Armando Elizondo Quiroga, armando.elizondo@insp.mx, Irma Sánchez Vargas, Saúl Lozano Fuentes, Karla Saavedra Rodríguez, Selene García Luna and William Black IV

In 2012, the Mexican Ministry of Health reported 50,000 cases of dengue. Veracruz State had the most cases and deaths associated with dengue in Mexico (25% of cases, 42 deaths). It was previously shown that *Aedes aegypti* in southern Veracruz is less competent to DENV infection. This study's objective was to determine the association of genes Dicer-2 exon 10 and Argonaute-2 exon 2 with vector competence to DENV-2 Jamaica 1409 among 7 collections of *Ae. aegypti* of Veracruz. Larvae and pupae were collected in August 2012 to obtain a representative sampling of eggs for analysis. The F2 generation was orally infected. Fourteen days post-infection, samples of saliva, midgut, head (salivary glands), and thorax/abdomen were collected. The infection titer was obtained by plaque assay. Percentages of infection were: midgut 20–70%, head 13– 60%, and saliva 0–26%. The Tuxpan collection demonstrated the least vector competence to DENV-2. The most susceptible was the isoform G (of the 1974 McClelland Score) from Martinez de la Torre. From every challenged mosquito, we extracted DNA, amplified Dicer2 and Argo-2 genes by PCR, obtained the phase and genetic distances, and checked for homozygotes and heterozygotes to confirm if homozygotes are susceptible and heterozygotes are refractory.

119 Molecular cloning, sequence analysis and gene expression of the circadian clock gene *Period* in *Culex quinquefasciatus* Say (Diptera: Culicidae)

Irám Pablo Rodríguez-Sánchez, iramrodriguez@gmail.com, Laura Mayela Montes-Rincón, Olga Karina Villanueva-Segura, Rafael González-Alvarez, Iván Delgado-Enciso, Adriana Elizabeth Flores-Suárez, Laura Elia Martínez-de-Villarreal and Gustavo Ponce-García

In prokaryotes and eukaryotes, the *Period* (*PER*) gene is important for the circadian clock that regulates some rhythmic biochemical, physiological, and behavioral processes. PER messenger amplification was done by RT-PCR from southern house mosquito, *Culex quinquefasciatus* Say, larvae collected at 3 locations in the State of Nuevo Leon, northeastern Mexico. The amplified products were cloned, sequenced, and compared/analyzed with those of other species of mosquitoes. A 3,093-bp amplified product was synthesized by PCR, which is the complete coding DNA sequence (3,027 bp) of *PER* encoding an open reading frame of 996 amino acid residues and much smaller than the orthologous in other mosquitoes analyzed. No other simultaneous amplification was visualized; thus, possible alternative splicings were discarded or at least not detected by the method. Because *PER* is a widely conserved gene in prokaryotes and eukaryotes, and maintaining colonies of southern house mosquitoes is relatively easy, determining the sequence of the gene in the mosquito provides the possibility of using the gene as a model and generating data on the role in the circadian clock.

120 Bioactivity of the essential oil of *Thymus vulgaris* L against *Stegomyia aegypti* (Diptera: Culicidae)

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Botanic insecticides are good alternative for the biologic control of insects that are vectors of diseases such as malaria, chikungunya, dengue, yellow fever, and several forms of encephalitis. In this work, the insecticide potential of the essential oil (EO) from *Thymus vulgaris* (Lamiaceae) against the dengue and chikungunya vector *Stegomyia aegypti* was evaluated. *Stegomyia aegypti* larvae between 3rd and 4th instar were used. Larvae were subjected to different EO concentrations (ranging from 25-47 ppm) during 24 h. Lethal concentrations (LC_{10}, LC_{20}, LC_{50} and LC_{90}) of EO were determined with Probit analysis. The effect of the EO on the enzymatic activity of acetylcholinesterase, glutathione S transferase, mixed function oxidases and several esterases (α-naphthyl-, β-naphthyl- and ρ-nitrophenol esterase) was analyzed, as these enzymes perform a number of crucial functions in insects and may be the target of the EO compounds. Enzymatic activities were spectrophotometrically determined using a microplate reader (Versa-max, Molecular Devices). The EO of *Thymus vulgaris* showed larvicidal activity with LC_{10}=26.52 ppm, LC_{20}=32.12 ppm, LC_{50}=46.29 ppm and LC_{90}=80.78 ppm. It was observed that from 26.5 ppm of EO the activity of the enzymes tested is affected. In conclusion, the EO of *T. vulgaris* has larvicidal activity and affects the metabolism of *Stg. aegypti*.

121 Effects of alkaloids derived from girgensohnine on mitochondrial bioenergetic of larvae *Stegomyia aegypti* (Diptera: Culicidae)

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The effects of AL47: 2-(3,4-dimethoxyphenyl)-2-(piperidin-1-yl) acetonitrile, AL48: 2-(3,4-dioxymethylenphenyl)-2-(piperidin-1-yl) acetonitrile, AL49: 2-(3,4- dimethoxyphenyl)-2-(pyrrolidin-1-yl) acetonitrile and AL50: 2-(3,4-dioxymethylenphenyl)-2-(pyrrolidin-1-yl) acetonitrile, 4 synthetic girgensohnine alkaloid analogs, were analyzed on *Stegomyia aegypti* larvae mitochondria. The enzymatic activities of NADH and succinate oxidases were determined by polarographic methods. Spectrophotometric methods were used for enzyme activities determination of NADH, succinate dehydrogenase, cytochrome c reductase and cytochrome c oxidase (at concentrations of 8 nM, 2 µM, 8 µM and 40µM). The compounds showed inhibitory activity of complex I, by reducing the activity of NADH dehydrogenase 20 to 25%, and NADH oxidase from 25 to 40%, the molecules AL49 AL50 showed a higher inhibitory activity. Succinate oxidase was inhibited by 20 to 40%, the compound AL49 showed the higher inhibition. Succinate dehydrogenase activity increased in the presence of the compounds (from 50 to 150%), suggesting a blocking of electrons transportation to oxygen that does not involve the succinate dehydrogenase enzyme. Succinate-cytochrome c reductase evaluation did not reveal effect of the compounds on the enzyme; unlike cytochrome c oxidase, which increased its activity by 25 to 100% in the presence of the alkaloids. Considering the results obtained for the different enzymes, an electron leak of the respiratory chain is suggested, favoring the formation of reactive oxygen species. These results contribute to the explanation of larvicidal effect that was previously reported for these compounds.

Disease/Vector Studies I

122 Incrimination of *Aedes* (*Stegomyia*) *hensilli* Farner as an epidemic vector of chikungunya virus on Yap Island, Federated States of Micronesia, 2013

Harry Savage, HMS1@cdc.gov, Jeremy Ledermann, Laurence Yug, Kristen Burkhalter, Maria Marfel and Thane Hancock

Two species of *Aedes (Stegomyia)* were collected in response to the first chikungunya virus (CHIKV) outbreak on Yap Island: the native species *Ae. hensilli* Farner and the introduced species *Ae. aegypti* (L.). Fourteen CHIKV positive mosquito pools were detected. Six pools were composed of female *Ae. hensilli*, 6 of female *Ae. aegypti*, 1 of male *Ae. hensilli*, and 1 contained female specimens identified as *Ae. (Stegomyia)* spp. Infection rates were not significantly different between female *Ae. hensilli* and *Ae. aegypti*. The occurrence of human cases in all areas of Yap Island and the greater number of sites that yielded virus from *Ae. hensilli*, combined with the ubiquitous distribution of this species, incriminates *Ae. hensilli* as the most important vector of CHIKV during the outbreak. Phylogenic analysis demonstrates that virus strains on Yap are members of the Asia lineage and closely related to strains currently circulating in the Caribbean.

123 Evaluation of novel dipstick assays for the detection of chikungunya virus in mosquitoes

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The recent recognition of chikungunya virus (CHIKV) in the Americas, with >730,000 cases diagnosed as of 26 September 2014, indicates the potential for CHIKV to cause an explosive epidemic. Because a licensed

vaccine is not available for CHIKV, our primary means of defense include mosquito control and methods to reduce mosquito contact. The ability to identify a pathogen rapidly under field conditions is an important part of both surveillance and disease suppression activities. Diagnostic assays, such as the VecTest dipstick for West Nile and St. Louis encephalitis viruses, are used to detect these viruses from infected mosquitoes and birds and have proven valuable in identifying areas of viral activity. To determine the efficacy of a similar assay to detect CHIKV, we evaluated a newly developed immuno-chromatographic (dipstick) assay. These dipsticks readily detected a single CHIKV-infected mosquito (including viruses from the East African, West African, and Asian lineages) in a pool containing up to 49 virus-negative specimens. We also tested these dipsticks with a variety of other alpha-, flavi-, and bunyaviruses to determine its specificity. Thus, the newly developed CHIKV dipsticks were highly specific and able to detect a single mosquito with a disseminated infection (i.e., a mosquito capable of transmitting CHIKV by bite) and should provide an additional tool in determining the presence of CHIKV in an area.

124 The North American Mosquito Project (NAMP) 2014 update: Using crowd sourcing and citizen science for continental scale mosquito collections

Lee Cohnstaedt, Lee.Cohnstaedt@ars.usda.gov and Elin Maki

The North American Mosquito Project was started in 2011 to collect disease vector mosquitoes on a continental scale. In the first year, mosquito control and public health professionals were used to collect mosquitoes. In 2012, the network was expanded to include citizen scientists to collect in areas not represented in the prior year. In total, 1,101 distinct mosquito collections were sent to the USDA in Manhattan, KS. The sample distributions covered the entire known distribution of *Culex tarsalis* and *Aedes vexans* in the continental United States. With this "snap-shot" of genetic material, population and phylogenetic analysis were done identifying the geographic origins of the species. Later in 2014, the network was again used to collect live samples from discrete genetic regions for vector competence studies. This presentation covers the formation of the network and the network uses. The collaborators' contributions were extraordinary and without their help, these projects are too costly in terms of time, man-power, and resources.

125 Elevated West Nile virus activity in California in 2014

Vicki Kramer, vicki.kramer@cdph.ca.gov and Tina Feiszli

West Nile virus (WNV) activity was elevated throughout California in 2014. Through October 2014, 654 human cases, including 22 fatalities, had been reported to the California Department of Public Health. This compares to a previous 5-year average of 206 human cases at this same time. Approximately 70% of the reported cases in 2014 had neuroinvasive disease. The number of WNV positive dead birds, mosquitoes, and sentinel chickens also exceeded the previous 5-year averages. The mosquito minimum infection rate statewide was the highest ever recorded in California. This presentation will include an update on WNV surveillance in California for 2014 and compare trends over the previous 10 yr. Factors influencing the level of West Nile virus activity in 2014 will be discussed, including California's drought conditions.

126 Invasive *Aedes* mosquitoes in California, 2011-2014

Vicki Kramer, vicki.kramer@cdph.ca.gov, Kerry Padgett, Melissa Yoshimizu, Renjie Hu and Marco Metzger

Invasive *Aedes* mosquitoes have recently been detected in California, including *Aedes albopictus* (2011), *Ae. aegypti* (2013), and *Ae. notoscriptus* (2014). Although *Ae. albopictus* and *Ae. aegypti* have been found previously in California, the recent detections represent populations that have become established. *Aedes albopictus* and *Ae. notoscriptus* have been detected in Los Angeles County, whereas *Ae. aegypti* have been found in 7 counties from several regions of the state. This presentation will provide information on the timeline, location, and modes of discovery. Results from genetic analyses of these populations will be presented. Ongoing surveillance and response activities by local vector control agencies and the California Department of Public Health will be described. Information on imported cases of dengue and chikungunya from 2011 through 2014 in California will also be presented.

127 Eastern equine encephalitis virus: reemergence and expansion in the northeastern United States

Theodore Andreadis, theodore.andreadis@ct.gov, Philip Armstrong and Goudarz Molaei

Eastern equine encephalitis (EEE) virus is the most deadly mosquito-borne pathogen in North America with an estimated human case fatality rate of 35 to 75%. EEE virus activity is most common near freshwater hardwood swamps in the Atlantic and Gulf Coast states and Great Lakes region, where the primary mosquito vector *Culiseta melanura* lays its eggs. Since the discovery of EEE virus in the 1930s, outbreaks in temperate regions have been sporadic, both temporally and spatially, highly focal, and largely unpredictable. However, over the last decade we have witnessed a sustained resurgence of EEE virus activity within long-standing foci in the northeastern US and unprecedented northward expansion into regions where the virus had been historically rare or previously unknown, including northern New England. This has resulted in severe disease in humans (51 cases and 20 fatalities) and domestic animals (213 cases). The factors

responsible for the reemergence of EEE virus are largely unknown but are likely complex reflecting ongoing changes in the ecology and epidemiology of this virus. The underlying factors associated with the introduction, amplification, and persistence of EEE virus in the region will be examined including 1) changes in land-use that increase human exposure near critical habitats; 2) vector mosquito abundance and distribution that drive viral amplification and spillover into human and equine populations; 3) species-specific mosquito-avian interactions that favor amplification, virus titers in primary and secondary mosquito vectors; and 4) genetic variation in regional EEE virus strains that provide evidence for local overwintering, evolution and extinction of EEE virus strains, with periodic reintroduction from southern sources.

128 Evaluation of aerial adulticide applications for *Aedes aegypti* and *Ae. albopictus* control in New Orleans, Louisiana

Sarah Michaels, srmichaels@nola.gov, Edward Foster, Brendan Carter, Laura Turpen, Cynthia Harrison, Princeton King, Claudia Riegel and Janet McAllister

Aedes aegypti and *Ae. albopictus* are widespread and abundant in urban neighborhoods of New Orleans, LA. Due to the potential for local transmission of chikungunya virus following the return of infected travelers, the New Orleans Mosquito Control Board evaluated existing control measures against these vector species. Dawn and dusk aerial applications of Dibrom® (naled) at 0.75 oz/acre were evaluated against field-collected caged mosquitoes. Cages were placed in open and sequestered locations in vegetation and under homes in 2 urban neighborhoods with high human density and abundant mosquito populations. Mortality of *Ae. aegypti* was 80.4 – 96.0% in sequestered locations and 86.4-97.9% in open locations and mortality of *Ae. albopictus* was 78.3– 95.8% in sequestered locations and 63.1-100% in open locations for dusk applications. Irregular results occurred during dawn applications likely due to shifting wind direction just prior to application. Differences in mortality can be explained by neighborhood and vegetation density as well as ability to deliver chemical effectively given urban obstructions affecting coverage. Repeated applications will likely be necessary to cause a significant reduction in natural populations. This study demonstrates that aerial adulticiding can be an effective component for *Aedes* species control and is an appropriate arboviral epidemic response.

129 Rapid emergence of *Ixodes scapularis* and associated tick-borne illnesses between 2006 and 2013 in Manitoba, Canada

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In 2006 a single established population of *Ixodes scapularis* (blacklegged tick) was identified in the southeastern corner of the province, along the border with Minnesota. Given this discovery, passive and active tick surveillance efforts, which had previously been discontinued due to low numbers and stable prevalence rates, were subsequently re-launched 2 yr later. These surveillance activities, a collaborative effort between Manitoba Health, the Public Health Agency of Canada and the University of Manitoba, have demonstrated a rapid and unexpected range expansion of blacklegged ticks, as well as increased prevalence of pathogens such as *Borrelia burgdorferi*, *Anaplasma phagocytophilum* and *Babesia microti* throughout much of the southern portion of the province over the past 8 yr. Established blacklegged tick populations are being discovered in clusters of forested or riparian areas found primarily in the eastern forest regions, along river valleys extending out into prairie habitat, and in some urban areas. This expansion has brought these ticks and their associated pathogens into closer proximity to more densely populated areas which has led to an increase in reports of illnesses associated with tick-borne disease such as Lyme disease, anaplasmosis and babesiosis. For example, the annual number of Lyme disease cases reported in Manitoba has increased from 5 in 2009 to 29 in 2013 and similar observations have been recorded for anaplasmosis. The ongoing range expansion and subsequent introduction of tick-borne pathogens has significant public health implications.

130 Eastern equine encephalitis in Escambia County, Florida

Kimberly Sargent, kssargent@co.escambia.fl.us and Bob Betts

As the residential and business communities move into closer proximity with the habitats of mosquito-borne disease vectors, the opportunities for transmission increase. The biological characteristics of the primary and secondary species found in the northwestern Florida Panhandle region are described. This review of eastern equine encephalitis disease impacts in Escambia County affecting susceptible humans and horses demonstrate that known sources, actions, inactions and precautionary measures may not prove to be enough to counter the development and emergence of the vectors as implicated by historical reference. A discussion on the local control measures and messages to the target communities may provide inspiration for other districts to take proactive steps in order to minimize the ecological impacts from these vector species.

Legislative and Regulatory I

131 An overview of the AMCA legislative and regulatory issues
Angela Beehler, angela@mosquitocontrol.org

Brief overview of legislative and regulatory issues that will make up the position papers for the Washington Conference. Presentation will mention steps the AMCA is taking to ensure pesticide regulations are based on sound science as opposed to fear and threat of litigation while addressing concerns about non-target impacts.

132 Mark your calendars for the 2015 Washington Conference
Roxanne Connelly, crr@ufl.edu

The AMCA Washington Conference provides an opportunity for Association members to receive updates on legislative and regulatory issues of greatest concern to mosquito control professionals and to become active participants in American democracy. After an afternoon of discussions and training, AMCA members proceed to Capitol Hill to meet with elected representatives and staff. Only by working together can we most effectively influence regulators and legislators. Please join us for the AMCA's 17th legislative conference, May 4-6, 2015 at the Hilton Crystal City hotel in Arlington, Virginia.

133 Mosquito control and the 114th United States Congress
Kam Quarles, kquarles@mwe.com

Update on the status of National Pollutant Discharge Elimination System (NPDES) legislation, our focus moving forward, and working with the current Congress.

134 Various L&R snapshots: 1) mosquito control on National Wildlife Refuges and a new USFWS Technical Handbook; 2) proliferation of adulticide barrier treatments; 3) continued CDC funding to the states for arbovirus-related work
William Meredith, William.Meredith@state.de.us

1) As a guidance document for USFWS staff, the Service is preparing a Technical Handbook for mosquito management on National Wildlife Refuges, with substantial potential to affect mosquito control practices not only on-refuge but off-refuge too -- the AMCA's concerns and roles in this initiative are discussed; 2) expanding use of adulticide barrier treatments in urban/suburban settings has specter for adverse non-target impacts that probably warrants more consideration for where things might be heading; 3) the CDC will continue as best as possible to provide annual federal funding to the states for arbovirus-related work, but the stability and amount for such funding remains in doubt and needs attention – more participation by AMCA members on the AMCA Federal Funding Subcommittee is also needed, not only for CDC grant issues but also for many other types of federal funding matters.

135 Mosquito control - endangered species and pollinator issues
Michael Hudon, m.hudon@irmosquito2.org

Endangered Species Act regulations and pollinator issues continue to foster conditions that result in mosquito control efforts being modified, restricted, or eliminated. This presentation will update you on some of these issues that do impact or in the future may impact mosquito control.

136 Regulation of our chemical control tools
Zane McCallister, grmcd1@acsol.net

This presentation will provide an introduction to the review process that all pesticides will go through in the near future. Malathion and chlorpyrifos products are currently under review, and we will discuss how you can provide valuable data. We encourage you to attend the second legislative session to focus on chemical control tools with opportunities for audience participation.

137 PESP becoming PestWISE
Gabrielle Sakolsky-Hoopes, gsakolsky@aol.com

AMCA has been a partner in the EPA's Pesticide Environmental Stewardship Program since 1997. In the winter 2014/2015, the program is being rebranded as PestWISE with a focus on risk reduction, education and training, and the economic benefits of using integrated pest management. The rebrand includes changes to membership levels and reporting requirements. AMCA and its partners continue our commitment to the program through these changes.

Operations

138 20+ years of collecting waste tires: one approach for control of dengue and chikungunya virus vectors

Douglas Carlson, doug.carlson@irmosquito2.org and Donald Shroyer

Since 1992, the Indian River Mosquito Control District has collected illegally disposed tires from fields, ditches and wooded areas throughout Indian River County (IRC), FL. Over this 22-yr period, nearly 100,000 tires (=1,000 tons) have been collected and transported to the county landfill. This ongoing project has helped achieve several objectives: reducing *Aedes aegypti* and *Ae. albopictus* larval habitat and intra-county dispersal along with helping to reduce litter in IRC. With the recent onset of dengue and chikungunya in southern Florida, this source reduction effort is taking on increasing importance in providing one means in which the mosquito vector populations are being reduced.

139 Unmanned aerial system integration in vector control - performance, payload, efficacy - how close are we to taking flight?

Bill Reynolds, breynolds@leateam.com and Piper Kimball

Unmanned aerial systems (UASs) have been the topic of many conversations. Brief experiences with UASs in mosquito control have occurred in the past; however, these were trial-based applications. In 2014, a UAS was designed based on aerial larvicide and adulticide requirements for vector control operations. Numerous operational applications were performed throughout California in 2014. This presentation will focus on the regulator integration of UASs into the National Airspace System (NAS), systematic application data, application precision and bioassay efficacy results. Detailed advantages of UASs in mosquito control operations will be discussed and demonstrated based on operational applications.

140 Computer methods to maximize larviciding

David DeMay, ddemay@keysmosquito.org

Targeting the application of larvicides is key to minimizing the need to adulticide. Three computer programs provide a nearly infallible search strategy to find mosquito development sites. Google Earth Maps provide the foundation for all 3 methods. The Growth Management division of Monroe County generated habitat maps throughout the county. Three significant habitats occur at elevations around 1 ft. Specifically, scrub mangroves, buttonwood, and mangroves are the optimal mosquito oviposition sites in the Florida Keys. Elevation contours can be created by the Sketchup computer program (sketchup.com) directly on Google Earth images and stored as .kmz files. Multiple layers can be generated subdividing contours. Elevations are verified referring back to the Google Earth computer program. A third method relies on an indicator plant. In the Florida Keys, saltwort nearly universally occurs alongside mosquito habitat. A specific html color code exists for saltwort. The specific html color code can be highlighted in any image using the computer program GIMP. Images from these 3 methods (County Habitat, Elevation Contour and Indicator Plant) can be superimposed or used individually to identify optimal mosquito oviposition sites. FKMCD has deployed entomological inspectors with specific map locations to monitor for inundation and breeding (fall 2014).

141 Surveillance data in 2014 and previous years: Tangipahoa Parish, Louisiana

Colby Colona, colby@tangimosquito.org

This presentation will discuss a brief background of Tangipahoa Parish mosquito control, trapping methods, and surveillance and disease data from 2008-2014.

142 Streamlining operations with data acquisition and display: MapVision, DropVision and SprayVision

James Clauson, jamesclauson@comcast.net

Data acquisition and display is central to an effective and efficient mosquito control operation. With the advent of new technology, many mosquito control districts are looking into the development and use of computers and GIS/GPS to streamline their operations. Beach Mosquito Control District (BMCD), located in Panama City Beach FL, has embraced this technology with the purchase of MapVision, DropVision and SprayVision. This data acquisition and display software has enabled BMCD to more effectively conduct larviciding and adulticiding operations. DropVision is software that is used for droplet analysis for adulticiding operations. SprayVision is software that is used to guide and track ground truck adulticiding. MapVision is software that ties it all together for data acquisition, tracking and display. In addition, required State reports and inventory controls are integrated and produced seamlessly. The ultimate goal of any technology should be to "make life easier" and the MapVision suite certainly does for Beach Mosquito Control District!

143 Sustainability: the use of bulk material and the elimination of forty-pound bags in the ground treatment process
Loren Lemke, llemke@mmcd.org

This presentation will show aspects of the Minnesota Metropolitan Mosquito Control District sustainability initiative and the processes used in accomplishing ground operations with *Bacillus thuringiensis israelensis* bulk corn cob materials.

144 Strategic use of pre-hatch larvicides can improve your mosquito control program
Mark Smith, mmcd_mes@mmcd.org

The Metropolitan Mosquito Control District (MMCD) continues to focus on improving its operations. District staff strives to leverage all processes, equipment, and control materials to maximize their effectiveness. By critically reviewing its operations, incorporating new ideas and focusing on organizational goals, MMCD has been able to continually increase the number of acres treated and better serve the citizens of our 7-county area.

145 An overview of county-wide mosquito control districts in southeast Texas
Kyle Flatt, kflatt@hcphes.org

There are 6 counties in southeast Texas that have mosquito control programs at the county and district level. These 6 districts, which were established in the 1950s and 1960s, serve approximately 5.3 million people. This presentation includes current capabilities, types of equipment, materials and methods utilized, as well as key species of concern.

146 The 100-year flood in Maricopa County, Arizona (2014) and the impact on our mosquito abatement program
Kirk Smith, ksmith@mail.maricopa.gov and John Townsend

In September 2014, 2 major tropical storms moved through Arizona from Baja California. As a result, widespread flooding occurred in locations throughout the state including Maricopa County (home of the greater Phoenix area). These storms were unique in that the entire county received significant rainfall (greater than 5 in.). Our stormwater systems were overwhelmed which resulted in several freeway systems shutting down due to flooding with many neighborhoods suffering similar consequences. In a matter of days, we received mosquito complaints throughout the county and the news media was relentless for information regarding our response. In the end over 10,000 complaints were processed. We will discuss the impact of this on our program including the dynamics of our mosquito species populations and incidence of West Nile virus.

147 Evaluation of truck-mounted application of VectoBac WDG against *Aedes albopictus* and other backyard breeders in Southern California
J. Wakoli Wekesa, wwekesa@sgvmosquito.org, Angela Brisco, Marta Tanaka, Antonio Bishop, Melvin Cook, Susanne Kluh and Kenn Fujioka

Low volume (LV) application of *Bacillus thuringiensis* subsp*. israelensis* in VectoBac WDG for control of *Ae. albopictus* in cryptic sources was evaluated. Three LV generators (Dyna Fog LV-8, Buffalo turbine [micronaire nasal] and London Fog 18-20 models) were used to evaluate the effectiveness of such approach for area-wide Asian tiger mosquito control. The impact of backpack sprayers, especially Stihl SR-450, as a supplemental sprayer to the Dyna Fog LV-8 shall be discussed.

Latin American Symposium III

148 Evaluation of organic and inorganic substrates for mass production of parasitic nematodes of mosquito larvae
Rafael Perez-Pacheco, rafaelperezpacheco@yahoo.com, Alicia Alonso-Ramos, Araceli Perez-Pablo, Gerardo Rodriguez_Ortiz and Edward Platzer

River sand is usually used as a substrate for nematode reproduction during mass production of parasitic nematodes of mosquito larvae, but the weight and the sand movement generated during transportation reduces yield of cultures by 80 to 90%. We evaluated organic and inorganic substrates (2 species of moss indigenous to Oaxaca and 1 species from the Republic of Chile, to replace sand as the culture media. Production of *Romanomermis culicivorax* and *R. iyengari* was examined in several experiments. 100 females and 100 males or 1 g of nematodes were added to each culture medium in polyethlene containers. After 4, 6, 8, 10 wk, cultures were flooded with distilled water to induce hatching and the number of pre-parasitic nematodes obtained in each experimental unit were determined. The cultures of 100 females and 100 males of *R. culicivorax* at 8 wk pre-parasitic nematode production in sand was 38,300 and in Chile moss 33,000. After 16 wk in sand, the yield was 32,000 and 64,000 in moss. In cultures with 1g at 8 wk in sand the

production was 53,200 and in moss 46,320. At 16 wk, the yield was 46,000 in sand, 40,000 in moss from Chile, and 42,000 in moss from Oaxaca. For cultures of 100 females and 100 males of *R. iyengari* in sand, 25,375 pre-parasitic nematodes were recorded and 16,625 in moss from Oaxaca. In the cultures with 1g of nematodes at 4 wk 237,750 nematodes in sand and 200,000 in moss from Chile. The pre-parasitic nematodes collected in the different experiments were used for the infection of larvae of *Culex quinquefasciatus* and the mosquito larvae were parasitized efficiently. This indicated that the substrates do not affect the infectivity capacity of nematodes.

149 Sectored brigade for high dengue transmission areas: an alternative operational program model for field control activities

Ildefonso Fernandez Salas, ildefonso.fernandez@insp.mx, Esteban Eduardo Diaz Gonzalez, Ewry Arvid Zarate Nahon, Iliana Rosalia Malo Garcia, Alejandro Gaitan Burns, Samanta Del Rio Galvan, Rosa Maria Sanchez Casas and Jose Genaro Ordoñez Gonzalez

Dengue outbreaks continue in most endemic countries despite intensive control efforts including development of new insecticides, bacterial and insect-growing regulators larvicides, potential transgenic mosquitoes, etc. An important gap between control measures and field applications (and results) deals with a weak and uncoordinated technicians field work. For instances, sometimes space-spraying using vehicle-mounted ULV generators is applied upon neighborhoods and days later larviciding activities are continued. Another common problem during field operations is to find closed households which remain untreated. Additionally, non-residential premises located within neighborhood such as small grocery stores, churches, schools, and vacant lots are not treated for *Aedes* mosquitoes. Clearly, combinations of these patchy application measures may explain in part the current failure of dengue operational program structure. We propose a new field technician structure; a sectored brigade focused on high transmission areas. The brigade is composed of 5 technicians, including 2 females to support house visits where only housewives are present. This new operational program has 3 innovative components to ensure its high effectiveness including: 1) exclusive assignment of the same brigade in a single sector with 1,600 homes for a year; 2) revisits closed and reluctant houses until achieving the control intervention; and 3) applying *in situ* most techniques for larval and adult control, community education, and support for active surveillance system to report suspected dengue fever cases.

150 A change in the perception of the population Quintana Roo, on the current situation of dengue, using the strategy of community involvement, Healthy Home "Casa salusable"

Marco Dominguez, dguezmarco@gmail.com, Pedro Mis, Ildefonso Fernandez, Jorge Esparza and Liliana Macias

Ensuring effective participation of the population is one of the major problems in the control of vector-borne diseases, and this question becomes vital to the effectiveness of the control measures in the specific case of dengue, as the vast majority of farms were located in residences or vicinity. From November 2012 to May 2013, in Quintana Roo, the community engagement strategy "Healthy Home" was implemented, which consisted of empowering people as to the key to the prevention and control of dengue, through the identification, removal and monitoring and evaluation of the risks of transmission by the presence of *Aedes aegypti* oviposition sites. During the "Healthy House" strategy, more than 1,000 people were added and trained of which 80% credited their homes due to their persistence in the elimination of mosquito oviposition sites and participation in health programs, such as possession of primers and pet vaccination. The role of these leaders was crucial to teach all family members to recognize the real risks of transmission of various diseases, including dengue and the added benefits of the strategy. He also worked with the whole structure of government State Health Services, some political leaders, who strengthened their participation the concept of community participation. Finally, we conclude that the state vector program requires a strong involvement in the community, supported by the municipalities and directed by the health services.

151 Mosquito records from Mexico VI: The mosquitoes of Hidalgo state (Diptera: Culicidae)

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In 2013, we conducted mosquito collection trips in the state of Hidalgo, Mexico with the objective to determine the biodiversity of culicid mosquitoes in all physiogeographic regions and sub-regions of state: Grand Folded Sierra with Huasteco Carso, and Neo-volcanic Axis with Sierras and Plains of Hidalgo and Queretaro states and Lakes and Volcanoes of Anahuac. The specimens were collected in both dry and rainy season. Adults were collected in different locations of state using CDC light traps, Shannon traps, Display light traps, while immature stages were collected in all water bodies available in the field using a dipper; larvae were later put into individual jars to let immatures develop to adult stages, thus getting the associated immature skins. All specimens were transported to the Parasitology Laboratory of Autonomous University Antonio Narro Laguna Unit in Torreon, Coahuila Mexico for mounting and identification. We obtained 2,521 specimens (657 adult female, 371 adult male, and 1,493 whole larvae and larval/pupal skins on microscope slides). In Hidalgo state inhabiting the 2 Culicidae subfamilies were: Anophelinae and

Culicinae; 6 tribes: Aedini, Culicini, Culisetini, Sabethini, Toxorhynchitini, and Uranotaeniini; 15 genera, 19 subgenera, and 44 species are recorded. Twenty-five nominal species were new records for Hidalgo state.

152 Molecular phylogeny of the subgenus *Melanoconion* of *Culex* (Diptera: Culicidae)
Carolina Torres-Gutierrez, aniloract@gmail.com, Carolina Torres-Gutierrez, Eduardo S. Bergo, Tatiane M. P. Oliveira, Kevin J. Emerson, Susan Greni and Maria Anice Mureb Sallum

Species of *Culex* (*Melanoconion*) Theobald are recognized as vectors of arboviruses such as Venezuelan equine encephalitis virus complex and West Nile virus. *Melanoconion* represents a diverse group widely distributed in the Americas. The species of this subgenus pose a real taxonomical challenge as the identification based on morphological traits is mainly based on the male genitalia that require well-trained skills for dissections of the structures. The current classification of the subgenus recognizes 2 major sections, *Melanoconion* and *Spissipes*, and several non-formal groupings within each. We investigated the phylogenetic relationships of groups of species within both sections for a total of 42 species, using fragments of 2 single-copy nuclear genes (CAD, HB) and the barcode fragment of COI mitochondrial gene. Results of the phylogenetic analyses employing maximum likelihood statistical methods strongly support both *Melanoconion* and *Spissipes* Sections as monophyletic groups. Furthermore, the *Spissipes* Section showed cladistics groupings consistent with morphological classification, suggesting molecular tools should be considered when studying the medically important species of *Spissipes* Section. As for *Melanoconion* Section, only a few monophyletic groups can be recognized that corroborate morphological classification, leading to a cause to reconsider the current taxonomic arrangement. We discuss the utility of the chosen genes as tools to infer phylogeny. Our results contribute to a better understanding of the classification of subgenus *Melanoconion* and a set of useful data to help with the identification tasks.

153 Rescheduled to 93a

Disease/Vector Studies II

154 Impact of transfluthrin on resistant mosquitoes
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Mosquito strains that show an increased tolerance to the chemical class of compounds with sodium channel modulator mode of action (pyrethroids, pyrethrins) are usually described as "pyrethroid resistant". Recent studies suggest that the level of metabolic resistance depends on the structure of the molecule and that structurally different compounds may still be effective. This study highlights the potential of transfluthrin to control resistant strains of mosquitoes. Treated surface contact bioassays, using different concentrations were performed on susceptible *Aedes aegypti*, east kdr-containing *Anopheles gambiae,* and metabolic resistant *Anopheles funestus* with the pyrethroids cypermethrin, cyfluthrin, deltamethrin, lambda-cyhalothrin, permethrin and the AI transfluthrin. While the discriminating dosages for pyrethroids with phenoxybenzyl-moiety have shown differences in terms of resistance between the 3 tested mosquito species, the structurally different transfluthrin with polyfluorobenzyl-moiety was unaffected by the detoxifying enzymes. This study suggests that transfluthrin has the potential to control metabolic resistant mosquitoes, because of its different structural formula to the tested pyrethroids which are used in vector control. The detoxifying enzymes of the *Anopheles funestus* FUMOZ-R mosquitoes were not able to degrade the transfluthrin molecule.

155 The house fly, *Musca domestica*: once again a vector of pathogens of public health significance
Jerome Hogsette, jerry.hogsette@ars.usda.gov

During the time of Walter Reed in Cuba and in subsequent years of the early 20th century, the house fly lost its image of being a good fly to have around, and became a dreaded vector of disease-causing organisms. In the US, it was dubbed the typhoid fly, and many research projects were launched to learn more about its management and control. After the end of the Second World War, efforts were made in the US and other developed countries to improve sanitation by closing sewer lines and disposing of garbage and hospital waste in a manner that kept them out of the environment. Although the image of the fly as a scourge of human health was still proliferated in society, many times with the help of companies selling fly control devices, the tendency of flies to be held responsible for large-scale transmission of pathogens seemed to decrease. Around the year 2000, publications began to appear in the literature indicating a renewed importance of the house fly as a vector of pathogens of public health significance, not only in the US, but in other countries as well. The US military considers flies to be a significant day-to-day pest that causes approximately 4% of personnel to be unable to perform their duties because of enteric disorders resulting from fly-transmitted pathogens. The purpose of this presentation is to increase the awareness among mosquito control experts that the house fly is more than just a nuisance. A brief history of pathogen transmission by house flies will be given, with emphasis on recent findings and current speculation about the possibility of house fly transmission of Ebola virus.

156 Findings from ecologic investigations of potential West Nile virus exposure sites, Orange County, California, during 2014

Laura Krueger, lkrueger@ocvcd.org, Robert Cummings, Tim Morgan, Kiet Nguyen and Amber Semrow

Orange County, CA, experienced an outbreak of West Nile virus (WNV) in 2014 that lead to an excess of 250 cases and 7 deaths (as of October 2014). The Orange County Vector Control District (OCVCD) responded to human cases in the affected areas with a neighborhood notification (door-to-door) campaign, inspection and treatment of residential larval habitats, and expanded mosquito surveillance and treatment of out-of-service swimming pools and underground storm drain systems. Case investigations and mosquito collections were conducted, on average, 3 wk following illness onset. Case investigations found a high prevalence of both non-existing and torn window screens on houses, a low prevalence of mosquito production, and few reports of biting mosquitoes surrounding case residences. Mosquito surveillance using gravid traps showed *Culex quinquefasciatus* as the predominate species in affected neighborhoods, with an average collection of 35 mosquitoes/trap night at or near potential exposure sites (range 0 – 299). Ninety-seven percent of potential exposure sites had a mosquito collection, with 23% of those sites having counts of more than 100 mosquitoes. As of October 2014, 29% of mosquito collections at or near potential exposure sites were WNV-positive by RT-PCR. Mosquito larvae were found around residential properties in <4% of door-to-door investigations, suggesting mosquito production was occurring somewhere other than backyard environments. Data collected from the ecologic investigations were used to modify OCVCD's Emergency Response Plan and direct future control activities.

157 An overview of a record outbreak year of West Nile virus in Orange County, California: Why did a well-established mosquito larvicidal program fail to prevent WNV amplification in urban *Culex quinquefasciatus*?

Robert Cummings, rcummings@ocvcd.org and Kiet Nguyen

While previous WNV activity in Orange County, CA, had oscillated on an apparent 4-yr cycle since its arrival in 2004, with recrudescence (2008 and 2012) and subsidence following large outbreaks, the county unexpectedly experienced its worst WNV outbreak on record. Approximately 300 infections with 7 deaths (as of November 2014) occurred in the county despite a well-established mosquito larvicidal program, public awareness campaigns, and early detection in mosquito and dead bird samples. Ten years of previous WNV activity had honed the Orange County Vector Control District's (OCVCD) larvicidal mosquito control program to focus its efforts spatially and temporally to disease prone areas during periods of likely virus activity. This presentation will discuss the significant challenges OCVCD faced when presented with expanding its disease suppression program to use adulticiding methods in a highly urbanized environment.

158 Epidemiology of West Nile virus in New York City: review of mosquito data, 2000 - 2014

Waheed Bajwa, wbajwa@health.nyc.gov

Since its first discovery in New York City (NYC) and in the Western Hemisphere in 1999, West Nile virus (WNV) has become firmly established in the city and the continental United States. During the last 16 yr, the city has experienced 5 outbreaks; the most significant was in 1999 when there were 59 human WNV cases, including 7 deaths. There was also, higher than average activity in 2002, 2003, 2010 and 2012. West Nile virus activity in mosquito populations of NYC is generally at its highest level from mid-July to early-September. This activity parallels with identification of most of the WNV human cases during the same time period. Based on virus isolation and the feeding behavior, *Culex pipiens, Cx. salinarius*, and *Cx. restuans* are the most likely vectors of WNV in the city. The abundance of *Cx. restuans* from late-May to July and isolations of WNV in late-June and early-July suggest that this species may play an important part in early season enzootic amplification of WNV. Its role as a bridge vector to humans is not expected to be significant as this species is mainly a bird feeder. *Culex pipiens* was abundant from early-June through September and *Cx. salinarius* in August and September. *Culex pipiens* was possibly involved in the early-season enzootic and both *Cx. pipiens* and *Cx. salinarius* in the late-season epizootic transmission of the virus. West Nile virus was frequently isolated from *Cx. pipiens* and *Cx. salinarius* in August and September, a period in which most human cases occurred. This concurrence and their abundance, and opportunistic feeding habits demonstrate that these species are the dominant vectors and possible bridge vectors of WNV infection to humans. We conclude that WNV activity in *Culex* mosquitoes substantially correlates with the risk of human infection that generally occurs mid-July through mid-October. As a result, the isolation of WNV from field collected *Culex* mosquitoes is an important and sensitive indicator of WNV activity in NYC.

159 Can the spread of *Aedes albopictus* be limited to current infested areas in Los Angeles County, California?

Angela Brisco, abrisco@sgvmosquito.org, "Albo Crew", J. Wakoli Wekesa and Kenn Fujioka

Since the discovery of the Asian tiger mosquito in September 2011 in the cities of El Monte and South El Monte, CA, the San Gabriel Valley Mosquito and Vector Control District has been conducting intensive surveillance to determine the intensity and extent of the infestation. Four years after its first discovery,

surveillance has shown a steady expansion of the area infested by *Aedes albopictus*. The spread, which was largely geometric rather than exponential for the first 3 yr, has reversed course to exponential spread in its fourth year. These findings have significant public health implications for California and the whole West Coast.

160 West Nile virus (WNV) surveillance in Harris County, Texas, 2002 – 2014
Martin Reyna Nava, mreyna@hcphes.org

Since West Nile virus' (WNV) arrival in Harris County, TX in 2002, 9,047 dead birds were tested with 1,644 confirmed in 37 species with blue jays comprising 29.4% of the collections. The percentage of WNV confirmed American crows was higher (64.5) compared to blue jays (43.8), albeit their collected number was 30 times lower. 2002 had the highest percentage of WNV confirmed dead birds (57), followed by the 2012 outbreak in Texas with 33. In addition, live bird sera collected from 23,147 specimens included 79 species with house sparrows, doves, and blue jays topping the list. Hemagglutination inhibition (HI) tests results for eastern equine encephalitis (EEE), Saint Louis encephalitis (SLE), and WNV were higher in after-hatch-year birds (AHY:76%) than in newly hatch- -year samples (HY:24%) overall. West Nile virus titers were higher in AHY samples whereas SLE titers were slightly higher in HY specimens. Furthermore, EEE resurfaced in 2014 after several years without detection. In addition, the number of positive mosquito pools totaled 5,689 from 14 species with *Culex quinquefasciatus* as the main vector for SLE and WNV. In 2014, 1,286 WNV-confirmed pools resulted with a minimum infection rate (MIR) of 3.8. The coverage distribution of confirmed mosquito pools in the county reached 87% compared to the next highest of 74% in 2006. Finally, the number of WNV and SLE human cases totaled 263 with 11 deaths. 2012 had the highest number of cases and deaths in Harris County during the Dallas outbreak.

161 *Aedes japonicus japonicus* (Theobald): A new emerging vector in northwest Florida?
Michael T Riles, michael.riles@comcast.net, James Clauson and John P. Smith

Aedes japonicus japonicus (Theobald 1901) is reported here for the first time in northwest Florida, USA. Six females and 2 males were observed and collected at separate sites using ovitraps during July 2014 and 1 male was collected at a separate site in a CDC light trap August 2014 in Bay County, FL. One female was reported in Okaloosa County, FL at 1 single site collected in a Mosquito Magnet X trap during August 2012. This specimen was misidentified then verified as *Ae. j. japonicus* in August 2014. The introduction and establishment of *Ae. j. japonicus* is significant concerning the possibility of a newly emerging vector within the state of Florida.

Legislative and Regulatory II

162 Chemical control toolbox - an open forum
Karl Malamud-Roam, kmr@aesop.rutgers.edu

Join mosquito control, members of industry, and federal regulators in a discussion about what you need from your chemical control toolbox. Are there enough chemical tools in your program's toolbox? Here is your chance to be heard! Topics of discussion include:

- The process of product registration review
- Date needs from the Environmental Protection Agency
- Changes that could appear on product labels
- Confusing or restrictive label language

Behavior/Biology

163 Growth and development of *Aedes aegypti* larvae at limiting food concentrations
Arieh Zaritsky, ariehzar@gmail.com, Tal Levi and Eitan Ben-Dov

Rates of growth and development were determined in individual *Aedes aegypti* larvae supplied with different concentrations of the defined nutritional food source Pharmamedia. At higher concentrations, rate of larval growth was faster, but the time required for 4th instar larvae to molt into the pupal stage was surprisingly extended and *vice versa* for lower food concentrations. The last larval stage pupates earlier in response to food deprivation, which displays striking degree of flexibility in the dynamics and timing of larval development. These opposite tendencies result in a constant time from hatching of an *Ae. aegypti* egg to pupation and up to adult eclosion at permissive food concentrations. Premature exhaustion of food supply might serve as a cue to initiate metamorphosis and therefore this phenomenon has ecological implications. It is likely that at least *Ae. aegypti* larvae respond to starvation by raising ecdysteroid titers and hence affects the timing of pupation. Manipulating food availability during larval growth and development may be

used to synchronize pupation and study pupal killing by delta-endotoxins of *Bacillus thuringiensis* subsp. *israelensis* ingested at the preceding 4th instar stage.

164 Floral preference of the malaria vector *Anopheles gambiae* (Diptera: Culicidae)

Mahmood Reza Nikbakhtzadeh, nik.nikbakht@gmail.com, John Terbot II, Philip Otienoburu and Woodbridge Foster

Mosquitoes of both sexes visit various plants to obtain enough sugar for their physiological needs. The main source of sugar for mosquitoes is floral and extrafloral nectaries. Semiochemicals assist mosquitoes in finding natural sources of sugar. To evaluate these olfactory preferences quantitatively, we used a 2-choice wind-tunnel olfactometer to measure the upwind orientation of *Anopheles gambiae* Giles, an important vector of malaria in equatorial Africa, toward odor plumes produced by 9 plant species common where this mosquito occurs. The primary purpose of this study was to rank the attractiveness of *An. gambiae* to a series of plant species, as measured by numerical response in a wind-tunnel olfactometer. A secondary objective was to identify the principal volatile organic compounds (VOCs) in the headspace of the most attractive plants to detect possible commonalities among their compositions.

Results presented here demonstrated that the volatiles of *Senna didymobotrya, Parthenium hysterophorus, S. occidentalis,* and *Lantana camara* were all attractive, compared to a control plant species. Chromatographic analysis of the headspace of attractive plants revealed a wide range of compounds, primarily terpenoids, e.g., enantiomers of caryophyllene, phellandrene, pinene, and ocimene. These chemicals are of interest because they can be exploited for malaria vector surveillance and control.

165 Evaluating the effects of mosquito adulticides on honey bees

Kristen Healy, khealy@lsu.edu, Jim Ottea, Todd Walker, Randy Vaeth, Frank Rinkevich and Joseph Margotta

The goal of this project has been to assess the risk of mosquito adulticides on honey bees by examining both toxicity and exposure. Our lab has been examining relative toxicity of the most commonly used adulticides, using Russian, Carniolan, and Italian honey bees. We have been assessing exposure in field experiments and during actual pesticide applications. The field experiments are being conducted in order to evaluate how control equipment and methodologies correlate with honey bee mortality. We will then use the data to create testable predictions that will be evaluated during actual pesticide applications in the field. The project will also assess long-term impact on honey bee colonies that were located inside and outside of mosquito control districts. By assessing these acute and long-term impacts, we can improve best management practices (BMPs) in mosquito control that minimize impacts on honey bees. We can also improve BMPs for beekeepers to ensure their bees are best protected during adulticide applications.

166 *Aedes pertinax*, a species newly recognized in the United States

Donald Shroyer, d.shroyer@irmosquito2.org, Bruce Harrison and Brian Byrd

Specimens of a mosquito new to the continental USA, *Aedes pertinax* Grabham, were retrospectively identified from 2 collections made in Indian River County, FL during 2011. This mosquito was initially thought to be a variant form of *Aedes tormentor*. Partial ribosomal DNA internal transcribed spacer (ITS2) sequences obtained from 2 topotypic *Ae. pertinax* collected in Jamaica in 1967 were identical to sequences from 2 Indian River County specimens collected in 2013. Routine mosquito surveillance in subsequent years has yielded more than 700 specimens appearing in more than 100 separate collections. The distribution of this mosquito in Florida and the United States is largely unknown, though it is probably a recent introduction. Recognition of the adult female and larva is hampered by morphological similarities to *Ae. atlanticus* and *Ae. tormentor*.

167 Species composition of the mosquito fauna of northwestern Uganda

John-Paul Mutebi, grv0@cdc.gov, Mary Crabtree, Jeremy Ledermann, Ann Powers, Barry Miller and Julius Lutwama

The mosquito species composition for a number of locations in Uganda was described during routine arboviral surveillance and outbreak investigations from the mid 1930s to the early 1970s. Recently, mosquito species composition for several locations in western Uganda, Zika Forest near Entebbe, and Mpigi in central Uganda has been described and/or updated. However, mosquitoes of northwestern Uganda have never been investigated despite the fact that West Nile virus (WNV) was first isolated from this region in 1937. In 2011 and 2012 the Uganda virus research institute, Entebbe (UVRIE) and the US Centers for Disease Control and Prevention (CDC) conducted arboviral surveillance at 4 locations: Chobe, Paraa, Sunguru and Rhino Camp in northwestern Uganda. In the course of this surveillance more than 75 different mosquito species were collected and 10 of these (*Aedes aegypti formosus*, *Anopheles funestus* group, *Culex annulioris*, *Cx. cinereus*, *Cx. decens* group, *Cx. duttoni*, *Cx. neavei*, *Cx. poicilipes*, *Cx. univittatus* and *Mansonia uniformis*) were collected at all sites. In this presentation, we will describe the species composition of the mosquito fauna at the 4 sites.

168 Ovitrap surveillance for *Aedes albopictus* in southeastern Massachusetts
Priscilla Matton, brismosqpc@comcast.net

The Bristol County Mosquito Control Project utilized oviposition cups for the first time in our Project's history to sample *Aedes albopictus* populations in southeastern Massachusetts. Ovitraps were used in locations were populations of *Ae. albopictus* have been collected since 2009. Traps were placed in the geographical locations of initial identification and just outside to establish range and species diversity throughout the season. Cups were sampled weekly using seed germination paper as an oviposition substrate. Eggs were counted, hatched and larvae reared to adults for identification. *Aedes japonicus*, *Ae. triseriatus* and *Ae. albopictus* were collected from 8 trapping locations from June to November during the 2014 season. Species composition will be examined and discussed.

169 Hump-shaped density-dependent regulation of mosquito oviposition site-selection by conspecific immature stages: theory, field test with *Aedes albopictus*, and a meta-analysis
Wasserberg Gideon, g_wasser@uncg.edu, Nicholas Bailes, Christopher Davis and Kim Yeoman

Oviposition site selection by gravid females is an important determinant of the distribution, abundance, and dynamics of dipteran hematophagous insects. The presence of conspecific immature stages in a potential oviposition site could indicate the suitability of that site but also indicates the potential for intraspecific competition. Our model suggests that the trade-off between these 2 opposing forces could result in a hump-shaped density-dependent relationship between oviposition rate and conspecific immature stage density with positive effects of aggregation prevailing at low densities and negative effect of intraspecific competition prevailing at higher densities. We field-tested these predictions with *Aedes albopictus* and evaluated if and how these relationships are affected by resource enrichment. Using oviposition cups containing varying numbers of conspecific larvae, we showed that the oviposition activity of *Ae. albopictus* first increases and then decreases with larvae number. Medium enrichment resulted in higher hatching rate, and demonstrated linear relations for the no-enrichment treatment where larvae density range was low and hump-shaped relationship for the enriched medium that had a wider larvae density range. Using pairs of oviposition cups, we showed that at low egg densities mosquitoes laid more eggs on substrates containing pre-existing eggs. However, at higher egg densities, mosquitoes laid more eggs on a virgin substrate. Based on our results and on a meta-analysis, we suggest that due to study design or methodological shortcomings the hump-shaped regulation model is often left undetected and that it is likely to be more common than currently thought.

170 Flying performance on different axial barriers in host seeking behavior of *Anopheles minimus*
Sungsit Sungvornyothin, sungsit.sun@mahidol.ac.th, Pitinat Sombuthoom and Ladawan Wasinpiyamongkol

Host seeking behavior of *Anopheles* females is important to both malaria epidemiology and prevention. Mosquito biting success increases both transmission rate of malaria and mosquito populations. Details of host seeking behavior are needed to interrupt biting success, which will decrease disease transmission and mosquito populations. Obstacle avoidance to different axial barriers during host-seeking, has been questioned—both in terms of understanding mosquito flight performance and identifying possible physical methods to integrate with other vector control measures. Experimental tunnels were attached with 3 different barrier models: 1) the X-axis, left-right, barrier; 2) the Y-axis, up-down, barrier; and 3) the combined XY-axis, diagonal, barrier. The experiment was done by putting mosquitoes into the starting side of tunnel and collecting the successful mosquitoes that reached the attractant chamber at the attracting side. The BG trap system™ was set in the chamber and released attractant. *Anopheles minimus* females were tested for 12 replications. The mosquitoes that reached the attracting side were collected every 10 min for 1 hr. Only 6.03% of mosquitoes successfully passed the diagonal retarded barriers compared to 12.64% of up-down, 31.31% of left-right and 50.62% of the control tunnels, respectively. The control and the left-right flying test groups showed significantly more success in seeking behavior than the diagonal and the up-down flying tested groups (Game-Howell multiple comparison, p-value = 0.007). It can be interpreted that *Anopheles minimus* performs more X-axis than Y-axis flying. The best barrier to retard host-seeking behavior should cause the mosquito to fly in a complex of axial flying.

171 Notes on cages, alternative culturing diets and methods for *Toxorhynchites rutilus septentrionalis*
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We have examined previously published *Toxorhynchites rutilus septentrionalis* lab rearing protocols for ways to upgrade methodologies to reduce rearing costs, larval duration, increase fecundity and fertility. We were able to determine the minimum cage size required for successful oviposition in wild sourced, lab-reared *Tx. rutilus septentrionalis* adults, and have designed a practical, inexpensive adult cage for mass rearing *Toxorhynchites*. Alternative diets and culturing methods were evaluated over the course of 1 yr in an effort to decrease larval developmental duration and reduce culture die offs.

Community-Based Control of Tick-Borne Diseases

172 The tool box for chemical control of ticks: current choices and some promising alternatives
Karl Malamud-Roam, kmr@aesop.rutgers.edu

The feasibility of community-based control of tick-borne diseases depends in large part on the availability of suitable chemical control agents, and this talk will introduce both current options and some promising new candidate materials and products. According to the National Pesticide Information Retrieval System (NPIRS), there are currently 1,717 end-use products registered by EPA for use against ticks, and these are based on 97 distinct active ingredients. The IR-4 Public Health Pesticides Program has built a database of these products and AIs, and the talk will summarize the product types and use patterns. Many of the products are repellents and pet products and will primarily be useful at the household/family scale, but there are a large variety of alternative toxicants that might be suitable for field-scale trials and possible adoption by vector control programs. In addition to existing products, promising novel approaches include pheromones, entomopathogenic fungi, botanical extracts, new synthetic toxicants and repellents, and new methods of dispersal.

173 Tick-borne diseases in humans the US
Charles Ben Beard, cbeard@cdc.gov

Lyme disease is the most common vector-borne illness in the US. In 2013, there were over 36,000 cases reported in the US making it the sixth most common of all reported diseases and conditions. Lyme disease cases have been increasing steadily over the last 20 yr both in numbers and in geographic distribution, a trend that is also observed for the other major reportable tick-borne illnesses. The cause for this increase is multifactorial, linked to a number of environmental and social changes. In contrast to mosquito-borne diseases, for which the responsibility for control is typically assumed by municipalities, tick control for prevention of Lyme and other tick-transmitted illnesses is shouldered primarily by individual homeowners. Lyme disease is frequently under-reported, with the actual number of cases in the US estimated at over 300,000 per year. The economic burden of illness in the US is great, with the cost of testing alone estimated to exceed over $490 million per year. There are also a number of complexities associated with prevention and control of tick-borne diseases, including opposition to the use of potential interventions including synthetic pesticides, deer removal, and human vaccines.

174 Tick-borne diseases in animals and USDA research on tick control
Andrew Li, Andrew.Li@ars.usda.gov

Tick-borne diseases represent a major threat to animal health in the United States. The cattle industry in the United States has benefited greatly from the continued USDA efforts through the Cattle Fever Tick Eradication Program in preventing the re-introduction of cattle ticks and associated pathogens that cause cattle fever disease (bovine babesiosis) from Mexico. However, frequent outbreaks of cattle ticks in the quarantine zone alone the US-Mexico border in Texas and the widespread presence of acaricide-resistant ticks both in Mexico highlight the risk of potential spreading of cattle ticks beyond the quarantined areas. Livestock production in the US is also threatened by a number of foreign tick-borne diseases, including Heartwater and African swine fever. The Gulf Coast tick (*Amblyomma maculatum*) is a known vector of pathogens that cause spotted fever rickettsiosis in humans and is also an efficient vector of pathogens that cause Heartwater. This tick species is expanding its range northward to reach Virginia currently. The soft tick *Ornithodoros* spp. are known vectors that transmit African swine fever virus in Africa and Europe, and a number of *Ornithodoros* spp. are present in the US. To mitigate the threat of tick-borne diseases to livestock production, USDA Agricultural Research Service (ARS) conducts research on tick-borne pathogens and tick control at several laboratories to develop novel control technologies. The 4-Poster Deer Treatment Bait Station developed by ARS scientists has been shown to be effective in reducing population density of both the lone star tick and blacklegged ticks in the northeast. New generation of anti-tick vaccines is being developed at the ARS laboratory in Texas. Research is also proposed to investigate the biology of *Ornithodoros* spp. and feral swine parasitism as risks for the emergence of African swine fever in the United States.

175 US Military efforts in tick surveillance and tick-borne disease prevention
Ellen Stromdahl, ellen.y.stromdahl.civ@mail.mil

US military involvement in tick surveillance is worldwide and long-term, because tick-borne disease is an occupational health risk for military personnel, and evaluation based upon infection prevalence in vectors represents disease risk more accurately than human case reports that are subject to misdiagnosis and travel history. Numerous Department of Defense (DOD) agencies cooperate to provide actionable evidence on tick species distribution and pathogen prevalence for installation commanders and military medical personnel. DOD entomological efforts are coordinated by the Armed Forces Pest Management Board. Laboratories at US Army Public Health Command and Naval Medical Research Center test ticks collected in their active and

passive surveillance programs. DOD epidemiologists from Armed Forces Health Surveillance Center track tick-borne disease case data and provide several publications. Walter Reed Biosystematics Unit and the Smithsonian Institution collaborate to offer TickMap.org, a geospatially referenced mapping program for tick species collection records and distribution models. In partnership with USDA, the DOD developed the permethrin-treated uniform, the most effective tool against ticks for military personnel. Continuing surveillance is warranted, because tick populations are rapidly increasing in size and range, novel tick-borne pathogens are being discovered, and increasingly sensitive pathogen detection methods are available; what we know today may not hold true tomorrow.

176 Federal recommendations for community control of tick-borne diseases
Susan Jennings, Jennings.susan@epa.gov

Integrated pest management (IPM) is a scientific strategy that uses pest surveillance and multiple control methods synergistically to reduce populations of target arthropod pests. The goal of IPM for prevention of tick-borne diseases is to reduce human illness and associated economic costs while minimizing potential environmental impacts. Discussions and consensus building between stakeholders, site users, and technical experts can help form an effective management strategy. Educational components help people reduce their risk of tick encounters, via improved awareness of landscape design and personal protection. Multiple US federal agencies currently share responsibility in addressing various aspects of tick-borne disease problems in the US. Through the coordination of efforts across these agencies, the US government has the opportunity to improve efficacy of control and reduce the risk from TBD. The Tick-borne Diseases Integrated Pest Management Workgroup (TBD IPM WG) was created to enhance communication and collaboration among US federal agencies involved in tick management.

Natural Compounds - What Can We Learn as an Industry? Can We Learn from Others?

177 Malaria vector control: back to the future!
Manuel Lluberas, lluberas@hdhudson.com

Malaria has been eradicated or reduced to a point where it is no longer a serious health or economic burden to a large number of countries. Almost without exception, eradication was reached by combining active vector population suppression methods and techniques and involving the local population to make their immediate environment less conducive to the proliferation of mosquito populations. More significantly, every country that achieved eradication did so more than half a century ago, long before the establishment of many of the current anti-malaria initiatives and without the benefit of modern technology or a vaccine. While much has been said about the expenditures related to malaria control and the funding shortfalls many programs face, there has been very little regarding their evaluation. Well into the twenty first century, public health entomologists continue to press the active mosquito control issue and wonder why the methods that eradicated malaria from so many countries—and has kept it out—continue to be overlooked and neglected by the agencies and organizations that promoted and implemented them so aptly early in the twentieth century. Examples of some of the past victories against malaria vectors are provided.

178 Controlling mosquito larvae naturally and effectively using entomopathogenic nematodes
Shea Dunifon, shea.porta@vegalab.com and Robyn Mucci

With increasing media exposures to diagnoses of chikungunya fever in Florida, Puerto Rico, and the US Virgin Islands, mosquito control products are garnering a lot of attention. An efficient means of preventing disease and pacifying public fears is to target mosquito development via natural and environmentally-sustainable products. Field studies conducted in South Florida using a biological control, entomopathogenic nematodes, demonstrated 100% mortality in mosquito larvae within 24 h after product application. We will be presenting the science behind this all-natural and environmentally-safe product and its potential for the US market. We will also present case studies of the product from Asia and Israel as this information will strengthen our case for the use of safer, natural and effective mosquito control products.

179 Greenspire Global - natural organic chemical products
Steve Knauss, smknauss@greenspireglobal.com

Environmentally friendly products Greenspire Global, Inc. develops, manufactures and distributes products for agricultural and commercial applications around the world, providing alternatives to harsh, toxic, and dangerous chemicals. Our products are effective tools for preventative and curative management programs of diseases, while promoting plant health and crop nutrition. We can help you reduce the use of chemicals that pollute soil, air and water. Our effective products allow you, season after season, to achieve your yields while enjoying healthier crops. Organic farming is a growing trend, driven by the awareness of the importance of maintaining yield volumes while observing practices that are environmentally friendly. Under

such premises, Greenspire Global® proudly offers its product portfolio, with highly effective tools for management of pests and diseases. Our products are characterized for being:

- Approved for use in organic agriculture
- Highly effective
- Broad spectrum
- Improves plant's defense mechanisms
- Applicable as standalone or tankmix

Including our products in your programs year around maximize each application event offering flexibility while remaining low risk for resistance generation. Our lineup is ideal for row crops, vegetables, fruiting trees, flowers and ornamentals. Consider our products for your farm whether for prevention or as a counteractive measure. A list of 5 of our products: Procidic, Safe Strike, Sunonic, Luxiant and Paramount.

180 Back to the future

Richard Howe, killmosquito01@gmail.com

This presentation is about how the mosquito control industry has ignored the science behind the need to dramatically reduce the size of the droplet that is necessary to control adult mosquitoes in an environmentally friendly way. As it stands, the industry is dumping over 99% of the insecticide on the ground that is not available for insect control because the droplets are too large to be useful in controlling mosquitoes.

The reason for this has to do with the use of spray systems designed to deposit insecticide on a crop. The term "space spraying" is all about treating the atmosphere for flying insect control. The droplets that do the work are true aerosol size droplets. These droplets are under 20 microns in size and do not deposit.

The Clean Water Act, if enforced, will effectively outlaw the aerial application of mosquito control insecticides unless the industry replaces the use of non-purpose built agricultural technology designed to deposit insecticide on a plant, to using true aerosol generators, thereby eliminating deposits into the aquatic environment.

181 Clean Water Act - what can we learn from other industries? Will mosquito control have a future?

Nicole Williams, williams@applicationdynamics.net and Tim Williams

The EPA Clean Water Act affects other industries besides mosquito control. Like public health, the agriculture industry has had to adopt different ways to comply with the EPA regulations. Both industries have major restrictions on the use of insecticides among other areas. We will review the different ways technology, natural compounds and advances in monitoring/applications have helped that industry to successfully comply with the Clean Water Act and still function successfully.

Surveillance and Control

182 Toxicological considerations of mosquito adulticide exposure to honey bees

Frank Rinkevich, fdr5@lsu.edu, Joseph Margotta, Jean Pittman, Bob Danka, Jim Ottea and Kristen Healy

Mosquito control relies heavily on truck and aerially mounted ULV application of pyrethroid and organophosphate insecticides. While these products are effective control agents, they are largely non-selective insecticides that may be a major concern to honey bee keepers. We used controlled laboratory studies to better understand the toxicological profile of these valuable tools for mosquito control on honey bees. The toxicity of the pyrethroids resmethrin (Scourge®), phenothrin (one component of Duet®), etofenprox (Zenivex®), and the organophophates naled (Dibrom®), and malathion (Fyfanon®) was assessed using 3 stocks of honey bees (Carniolan, Italian, and Russian). Among the pyrethroids, resmethrin was significantly more toxic to honey bees than phenothrin or etofenprox. There was variation in resmethrin sensitivity across stocks of bees with Italian bees being the most sensitive. The organophosphate naled was slightly less toxic than resmethrin across all stocks of bees, but significantly more toxic than phenothrin or etofenprox. Malathion was only half as toxic as naled and the least toxic overall. While organophosphates were generally less toxic than pyrethroids to honey bees, the dose range between 0 to 100% mortality was significantly more narrow than to pyrethroids. Sensitivity to phenothrin and naled varied with age with older bees being less sensitive to phenothrin, while older bees became more sensitive to naled. These findings provide considerations for which insecticide should be used to reduce toxic hazard where honey bees are of concern as well as information that could be used when relaying information to bee keepers.

183 Withdrawn

184 Where source reduction is not an option III
Francis Bosch, franb@co.williams.nd.us

Seven years after starting with a new Board, crew, and Director, the Williston Vector Control District is now poised to become a county-wide District. Collaboration with other government entities, college internship programs, and "missionary tactics" designed to increase retention have all augmented the District's efforts. While remaining anchored in the basics of mosquito control, the District has also had to implement unorthodox methods to keep up with the rapidly changing political and physical landscape of the oil boom area.

185 Comparative trapping efficiency of three gravid mosquito traps in field and semi-field habitats in northeastern Florida
James Cilek, james.cilek@med.navy.mil, Jennifer Wright and Jeremy Anderson

Trapping efficiency of the CDC gravid trap, BioGents gravid trap (GAT), and the CDC autocidal gravid trap (AGO) were compared. All traps were baited with a live oak/pine leaf litter infusion. Five backyards were chosen for field trials. All 3 traps were placed in each backyard at least 7 m apart. A 3 X 3 Latin square design was used for daily trap rotation/wk and mosquito collections obtained at 24 h. Gravid *Culex quinquefasciatus* were primarily collected from each trap. Using the CDC gravid trap as the standard, the efficiency of the GAT and AGO to collect this species will be presented. In addition, similar semi-field efficiency trials conducted in a walk-in screened cage using laboratory-reared *Aedes aegypti* and *Ae. albopictus* will be presented.

186 Modifications to the Prokopac aspirator for anthropophilic mosquito collections
Kevin Caillouet, caillouet@stpmad.org, Mike Francis, Kevin Lowrie and Charles Palmisano

While collection of human host-seeking mosquitoes can be tedious and inefficient, counts of landing mosquitoes are often not species-specific and mosquitoes are not retained for arboviral testing. We modified the Prokopac aspirator for "human-swarming" collections to supplement human landing counts for chikungunya virus vectors and other anthropophilic species. Modifications were intended to make the aspirator a lightweight handheld collection device. A custom aluminum welded battery bracket and handle with a 2.5 Ah battery allow the device to be used with one hand for up to 25 min per charge. The modified aspirator can also be used to collect adult mosquitoes from subterranean rainwater extension pipes.

187 Evaluation of relative effects of carbohydrate sources in yeast-fermentation CO_2 generators on mosquito surveillance
Robert L. Aldridge, robert.aldridge@ars.usda.gov, Seth C. Britch and Kenneth J. Linthicum

Mosquito surveillance in remote areas with limited access to canisters of CO_2 or dry ice will benefit from an effective alternative CO_2 source, such as the natural production of CO_2 from yeast fermentation of a carbohydrate such as sugar (sucrose). In this study, we explore the efficacy of CO_2 production from yeast fermentation of sucrose and 2 sucrose alternatives, molasses and honey, by comparing mosquito collections from CDC light traps baited with these 3 CO_2 sources and a dry ice standard in field conditions. Initial results have positively shown yeast generated CO_2 does attract mosquitoes to a trap, regardless of the carbohydrate source. Early results indicate no significant difference in capture rates of mosquitoes using CO_2 generated through yeast fermentation among all 3 carbohydrate sources.

188 Epidemiology of West Nile virus in New York City - review of mosquito data, 2000 - 2014
Howard Epstein, howard.epstein@emdmillipore.com and John DeBlasi

Since its first discovery in New York City (City or NYC) and in the Western Hemisphere in 1999, West Nile virus (WNV) has become firmly established in the city and the continental United States. During the last 16 yr, the city has experienced 5 outbreaks; the most significant was in 1999 when there were 59 human WNV cases, including 7 deaths. There was also, higher than average activity in 2002, 2003, 2010 and 2012. WNV activity in mosquito populations of NYC is generally at its highest level from mid-July to early-September. This activity parallels with identification of most of the WNV human cases during the same time period. Based on virus isolation and the feeding behavior, *Culex pipiens; Cx. salinarius*; and *Cx. restuans* are the most likely vectors of WNV in the City. The abundance of *Cx. restuans* from late-May to July and isolations of WNV in late-June and early-July suggest that this species may play an important part in early season enzootic amplification of WNV. Its role as a bridge vector to humans is not expected to be significant as this species is mainly a bird feeder. *Culex pipiens* was abundant from early-June through September and *Cx. salinarius* in August and September. *Culex pipiens* was possibly involved in the early-season enzootic and both *Cx. pipiens* and *Cx. salinarius* in the late-season epizootic transmission of the virus. WNV was frequently isolated from *Cx. pipiens* and *Cx. salinarius* in August and September, a period in which most human cases occurred. This concurrence and their abundance, and opportunistic feeding habits demonstrate that these species are the dominant vectors and possible bridge vectors of WN virus infection to humans. We conclude that WNV activity in *Culex* mosquitoes substantially correlates with the risk of

human infection that generally occurs mid-July through mid-October. As a result, the isolation of WNV from field collected *Culex* mosquitoes is an important and sensitive indicator of WNV activity in New York City.

189 Managing development and mosquitoes: a balancing act with examples from Australia
Patrick Dwyer, patrick.dwyer@dpi.nsw.gov.au, Jon Knight and Pat Dale

Population and its expansion is concentrated along the coastal areas of Australia. As a result greenfield sites are developed, often in close proximity to mosquito habitats. This reduces the quality of life for residents and/or involves large costs in mosquito management. Although urban designs that incorporate buffers between development and coastal wetlands can reduce the impact of some common nuisance mosquitoes, buffers do not address the risk from the saltwater mosquito, *Aedes vigilax* a major vector of Ross River and Barmah Forest viruses in Australia. That is because this species can travel up to 50 km to find a blood meal, especially when wind assisted. Control is most effective when targeted at the larval stage and larvicides may be routinely and repeatedly applied, at significant cost. Their efficacy can, however, be significantly reduced in a range of situations including adverse weather, where the pH is low (may be <3) or where there is dense canopy cover. Alternative habitat based control measures such as establishing runnels (a form of OMWM) within saltmarsh, and rehabilitation of mangrove wetlands to restore tidal flushing, can reduce mosquito production in source areas while retaining or improving the delivery of ecosystem services. Here we highlight with examples habitat based mosquito control measures together with urban planning and ongoing management of peri-urban areas.

190 Using Xeripave Tray Inserts to reduce mosquito breeding
Jennifer Henke, jhenke@cvmvcd.org and Gabriela Perezchica-Harvey

Physical control is an important aspect of integrated vector management programs and includes using methods that prevent adult mosquitoes from accessing standing water. The Coachella Valley Mosquito and Vector Control District coordinated the installation of Xeripave Tray Inserts at 20 catch basins on a single street in Cathedral City to determine if they would have an impact on the mosquito population in the neighborhood. The catch basins were siphons, meant to hold standing water but were not connected to a retention basin by an outlet pipe like traditional catch basins. Thus, the Xeripave Tray Inserts had the potential to completely eliminate the ability of mosquitoes to access standing water if the surface of the water was kept below the level of the paver trays. The pervious pavers allowed water to pass through but keep anything larger than 30 microns trapped at the surface. Xeripave Tray Inserts were installed 7 in. below the level of the street to accommodate the metal grates already present. Mosquito treatments were monitored before and after installation and were only made if mosquito larvae were found. Adult mosquitoes were also monitored using a gravid trap at the south end of the street. From June to October, the water temperature above the Xeripave Tray Inserts was warmer than it had been the previous year. Mosquito treatments were reduced by 64%, and the adult mosquito population was reduced by 35% over the 18-week period. Water stood above the paver trays, where mosquito production did occur. If water conservation methods are employed in the neighborhood, it is possible that the standing water would drop below the surface of the paver trays, further reducing mosquito treatments and the risk of West Nile virus transmission to residents.

191 Surveillance and control of exotic mosquitoes in Germany
Norbert Becker, norbertfbecker@web.de

Globalization resulting in increased human mobility and international trade is responsible for the quick spread of pathogens and neozoa such as container-ovipositing *Aedes/Ochlerotatus* mosquitoes. Within the invasive mosquito species, the Asian tiger mosquito *Aedes albopictus* (Skuse) plays an outstanding role because of its biological features. Since its introduction to Italy 1990 through the international used tire trade, the Asian tiger mosquito is spreading along the Mediterranean Sea, particularly by public and private traffic. In recent years findings of *Ae. albopictus* accumulate in more northern parts of Europe. Therefore, a national-wide surveillance program was launched by the German Federal Ministry for Environment, Nature Conservation and Nuclear Safety (BMU) under the leadership of the Bernhard-Nocht-Institute for Tropical Medicine (BNI). Highways connecting southwest Germany to regions in southern Europe where *Ae. albopictus* is already established, were specifically investigated by means of ovitraps and BG-sentinel traps to detect possible ports of entry of exotic mosquito species as well as to assess the risk of their establishment in Germany. In the last years, we found an increasing number of adults and egg batches of *Ae. albopictus* and a dramatic spread of *Ochlerotatus japonicus* in southwest Germany. The control of container inhabiting mosquitoes is mainly based on environmental management and the use of *Bti*-tablets in the frame of community participation. Since 2013 copper, a mosquitocidal metal, successfully deters mosquito oviposition.

Push-Pull-Killing Strategy for Adult Mosquito Control I

192 Future prospects for using new repellent developments in a push-pull strategy
Daniel Strickman, dan.strickman@gatesfoundation.org

Push-pull strategies have been shown to be effective against some insects. There could be many advantages to such a strategy, including better effectiveness, the ability to selectively eliminate specific parts of a vector population, and reduction in the amount of pesticide required. The biggest limitation in applying this strategy to human-biting mosquitoes has been the lack of practical chemical tools for both repellence and attraction. Recent study of the physiological basis of attraction and repellence has opened the door to completely new methods for chemical discovery. Chemicals designed to affect particular receptors should be active at much lower concentrations than repellent active ingredients like DEET. Volatile pyrethroids like metofluthrin and transfluthrin offer some possibilities that are available now. Solid repellents, like indole, might be useful in a push-pull strategy. Further, in the future we may be able to integrate chemically based repellents with physical repellents like infrared light and unnatural magnetic fields. An efficient system might appeal to the public's consistent desire for trap-based control techniques.

193 Topical repellents used as push strategy against mosquitoes
Mustapha Debboun, mdebboun@hcphes.org

Topical insect repellents are usually the first line of defense and give every individual person control over exposure to biting mosquitoes. Use of EPA approved and registered insect repellents to protect personnel from biting mosquitoes containing the active ingredients DEET, picaridin, oil of lemon eucalyptus, and IR3535 will be discussed.

194 Spatial repellents for control of vector-borne disease
Nicole Achee, nachee@nd.edu, John Gimnig, Mary Hamel, Sarah Moore, Thomas Scott, Jennifer Stevenson, Din Syafruddin and Neil Lobo

As goals of malaria elimination and eradication are put forth, and the scope of dengue continues to grow, further optimization of current products and/or new paradigms and tools will be required to meet public health demands. Spatial repellent (SR) products have gained renewed attention as one such paradigm. This presentation will outline key components of a critical path of development for inclusion of spatial repellents in disease control programs and introduce the framework of a recently funded 5-yr multicenter program intended to demonstrate and quantify the protective efficacy (PE) of spatial repellent products in reducing the incidence of malaria and/or dengue infections in human cohorts. Secondary objectives of the program will also be discussed as they relate to key issues to the optimization and application of SR products for public health such as efficacy against varied vector bionomic characteristics, within a range of baseline transmission intensity levels, and community effect as a function of diversion. Findings from the project will be available to global health authorities to support decision-making for inclusion of a SR strategy for disease control.

195 Chemicals that disrupt host-seeking in insects
Ulrich Bernier, uli.bernier@ars.usda.gov

Certain chemicals, e.g., linalool and nepetalactone, the primary component of catnip oil, have been reported to repel and inhibit the host-seeking of the yellow fever mosquito, *Aedes aegypti* (L.). More recently, chemicals have been identified which interfere with CO_2 reception in mosquitoes, and these chemicals are being explored as a means of personal protection through hiding hosts from mosquitoes. In our studies, we focus on the discovery of compounds that inhibit the attraction to human odors. These attraction-inhibitors impair the ability of insects to respond to kairomones that they would normally use as cues to locate the host.

A triple cage olfactometer was used to assess the attraction-inhibition level of compounds identified initially in human skin emanations. Additional compounds of similar structure were obtained commercially or synthesized to develop a better understanding of the class of chemicals which produce this inhibition effect. These 5-7 membered ring compounds with 1 or 2 nitrogens in the ring have been shown to impair host-seeking in female *Ae. aegypti*, *Anopheles albimanus* Weidemann, and *Phlebtomus papatasi* Scopoli. We have compared these chemicals to catnip oil (likely the best natural compound attraction-inhibitor) and to N,N-diethyl-3-methylbenzamide (DEET) . Many of these cyclic inhibitors are superior to catnip oil and DEET when studied in the olfactometer. Our most recent studies demonstrate that when caged adult female mosquitoes are exposed to these compounds for 10 min, the majority of the exposed females do not attempt to seek out the hand of a human host when the hand is inserted in the cage.

196 ThermaICell mosquito repellents kill several species of adult mosquitoes
Chris Bibbs, bibbsamcd@bellsouth.net and Rui-De Xue

ThermaCELL MR-GJ mosquito repellent was evaluated in semi-field trials (2,337 cubic foot outdoor space) against several species of mosquitoes. The repellent devices showed effectiveness with high KD and mortality, such as *Aedes atlanticus* with 98% mortality, *Ae. taeniorhynchus* with 84% mortality, *Psorophora ferox* with 97% mortality, and *Ps. columbiae* with 96% mortality. The device may have further practical application in domestic settings to help combat dengue and chikungunya virus exposures.

Recent Developments in Mosquito Surveillance and Trapping Techniques I

197 Use of passive traps for the surveillance and control of dengue vectors
Roberto Barrera, rbarrera@cdc.gov

There are a number of passive devices (i.e., do not use a power source) to collect or trap specimens of dengue vectors such as ovitraps, larvitraps, resting boxes, emergence traps, sticky surfaces and traps, funnel traps, and insecticidal traps. The most common baits used in passive traps are water, plant infusions, or synthetic lures that attract gravid female mosquitoes. Resting boxes and sticky surfaces attract females in various physiological states. No passive trap exists specifically to attract and collect blood-seeking *Aedes* females. The passive device most commonly used for the surveillance of dengue vectors is the ovitrap. Sticky gravid traps are being used to a more limited extent, but it is anticipated that they will be more frequently used in the near future. One study showed a significant association between the numbers of *Aedes aegypti* females in CDC sticky gravid-ovitraps (AGO traps) and BG-Sentinel traps. Emergence traps and sticky surfaces are useful for monitoring mosquitoes emerging from underground aquatic habitats such as wells and septic tanks. Passive devices have seen limited application for vector control purposes, such as ovitraps that use larvicides or contact insecticides (lethal ovitraps), sticky gravid traps, and pyriproxyfen/pathogen auto-dissemination stations. Recent investigations have shown that AGO traps caused significant and sustained reductions of *Ae. aegypti* populations.

198 Efficacy of carbon dioxide and synthetic lure-baited traps for surveillance of *Stegomyia* (Diptera: Culicidae) in North-Florida
Peter Obenauer, peter.obenauer@med.navy.mil, Peter Nunn, A. Richardson and J. Harwood

The recent introduction of chikungunya into Florida, as well as the resurgence of localized dengue transmitted cases within the United States, demonstrates the necessity for a reliable, durable, and light-weight mosquito surveillance trap to target these daytime-feeding vectors. We evaluated the capture efficiency of CO_2-baited and synthetic lure-baited BG-Sentinel 2 (BGS-2)™, BG Mosquito Tiger (BGM) and Zumba™ traps, and the Centers for Disease Control and Prevention (CDC) light trap for collecting *Aedes albopictus* (Skuse) and *Ae. aegypti* (L.) in 4 suburban neighborhoods and in a semi-field enclosure from May to August 2014. A total of 3,375 mosquitoes, representing 23 species from 10 genera were collected. Significantly more *Ae. albopictus* and *Ae. aegypti* were collected in the BGS-2 and BGM compared to the Zumba™ and CDC traps ($P < 0.001$). Durability and efficacy of the traps will be discussed.

199 Next generation BGS trap? Field comparison of Biogents Sentinel Trap 1.0 and 2.0 for monitoring *Aedes albopictus*
Isik Unlu, iunlu@mercercounty.org, Kim Klingler, Nick Indelicato and Ari Faraji

Aedes albopictus is a major nuisance pest because of its anthropophily and aggressive daytime biting behavior. It is also a public health concern because of the role it plays in the transmission of diseases caused by chikungunya and dengue viruses. Therefore, there is a growing demand for effective surveillance tools for this species. Biogents Sentinel (BGS) traps (Biogents AG, Regensburg, Germany) have proven to be an effective tool for surveillance of this species, but to increase the field stability of this trap, an upgraded version will soon become commercially available. In this study, we compared the performance of the new generation BGS traps to the original BGS traps that are widely in operational use. We deployed different combinations of BGS traps (with and without CO_2) in central New Jersey. The original BGS traps with CO_2 collected more *Ae. albopictus* than traps without CO_2 ($P = 0.0006$). The original traps also collected more *Ae. albopictus* when compared to the newer BGS traps with ($P=0.0001$) or without CO_2 ($P=0.00001$). The newer BGS traps collected more *Ae. albopictus* when CO_2 was added when compared to the same traps without CO_2 ($P = 0.017$).

200 A comparison of five different lure types used with the BG Sentinel 2.0 surveillance trap
Charles Abadam, cabadam@suffolkva.org, Karen Akaratovic and Jay Kiser

The BG-Sentinel trap is an important adult mosquito surveillance tool. In Suffolk, VA this trap has proven to be highly effective at monitoring *Aedes albopictus* and *Culiseta melanura*, primary vectors for West Nile virus and eastern equine encephalitis, respectively. This study compares 5 different lure types used with the

BG Sentinel trap model 2.0 by evaluating the adult mosquito population caught in each. The 5 lure types evaluated include the original 3-component BG mesh lure, the 3-component BG lure cartridge, a 4-component BG lure cartridge, a 1-component BG lure cartridge, and a control with no lure. Traps were set in a wooded area with a large tire pile adjacent to a children's playground, basketball court, and residential neighborhood. Using a Latin square design each trap/lure combination occupied 5 different sites 3 times each, producing 15 replicates. Results showed that traps baited with no lure caught the most mosquitoes overall and was comprised of *Cs. melanura* (59%), *Ae. albopictus* (9%), and *Psorophora ferox* (21%). *Aedes albopictus* comprised over half of the overall catch in traps baited with the 4-component BG lure cartridge (58%), the 3-component BG mesh lure (51%) and the 3-component BG lure cartridge (63%).

201 Trials and tribulations of troubleshooting Biogents® BG-Sentinel
Jay Kiser, jkiser@suffolkva.us, Charles Abadam and Karen Akaratovic

Over the last decade BG-Sentinel traps have been essential components in many adult mosquito surveillance programs around the world. During these years, the design of these traps has remained relatively unchanged with the exception of slight variations in building materials. However, the recent creation of the new BG-Sentinel trap, model 2.0, resulted in modifications of both the design and materials of the older BG models. Through a series of comparison studies and observations, various advantages and disadvantages were found to be associated with many of these modifications. Changes in cover material, funnel design, and exterior coloration among other components may be regarded as useful or problematic depending on how the equipment is used. The utilization of rain protection and proper transport of traps and catch chambers can greatly affect the value of these changes. The benefit of altering funnel design and exterior coloration is the flexibility to switch between model 2.0 and older trap models. In contrast, the non-removable PVC coated polyester fabric used in the manufacture of the outer body and inner core seems to have an adverse effect on mosquito capture rates. For example, 2 studies conducted, 1 in Suffolk, VA, USA and 1 in Regensburg, Germany, showed lower capture rates with the fabric used in this trap model than in older models without. Further investigations will be performed to evaluate the effectiveness of existing and new modifications.

202 Observations on the attractancy of *Aedes albopictus* to potential oviposition and nectar sites
Dan Kline, Dan.Kline@ars.usda.gov

Observations were made on the attractancy of *Aedes albopictus* to various artificial oviposition sites in 2 backyards. In 1 backyard, oviposition activity was compared between heated and unheated buckets. In the second backyard, seasonal oviposition activity was recorded in a potato chip bag, a "Tiger Tube," a green cemetery vase, and the standard "Little Black Jar". Olfactometer studies were conducted to determine the attractancy of several flowering plants to both sexes of *Ae. albopictus*.

Spinosad for Mosquito Control: From the Laboratory to the Field I

203 *Saccharopolyspora spinosa*, spinosyns, spinosad, and spinosoids
Tianyun (Steven) Su, tsu@wvmvcd.org and Min-Lee Cheng

Saccharopolyspora spinosa Mertz and Yao, an aerobic, gram-positive, non-acid-fast actinomycetes with fragmenting substrate mycelium was isolated in 1985 from soil in Virgin Islands. Spinosyns occur in over 20 natural forms, which have a generalized structure consisting of a unique tetracyclic ring system attached to an amino sugar (D-forosamine) and a neutral sugar (tri-*O*-methyl-L-rhamnose). Spinosad contains a mixture of spinosyn A (C41H65NO10) and D (C42H67NO10) in an approximately 17:3 ratio. Over 200 semi-synthetic spinosoids, such as spinetoram (a mixture of spinosyn J and L), have been produced in the laboratory. As a new class of polyketide-macrolide insecticides, spinosad has pesticidal activity after ingestion and cuticle absorption against a broad spectrum of susceptible insect species, acting at the nicotinic acetylcholine receptors (nAChR) and the γ-aminobutyric acid receptors and causing rapid and over excitation of the insect nervous system, leading to the mortality of the target species. Spinetoram is more active than spinosad against mosquitoes. There is a cross resistance to spinetoram in spinosad-resistant mosquitoes.

204 Toxicity of spinosad to *Culex quinquefasciatus* and three, non-target species of insects
Jim Ottea, jottea@lsu.edu and Owen Jones

Spinosad shows promise for control of larval mosquitoes, but there have been few studies on the effects of this insecticide on non-target aquatic insects. We surveyed a neighborhood pond to find a representative species of mosquito (*Culex quinquefasciatus*), as well as 3, common aquatic insects (a damselfly, a dragonfly, and a mayfly) with which to examine susceptibility to spinosad. Dosage mortality relationships were examined between Natular™, an EC formulation of spinosad, and larvae from a reference susceptible (Sebring) strain, and mosquitoes captured at the study site. Two concentrations of Natular™ were used to measure the susceptibility of the non-target insects: the LC_{50} calculated for field-collected mosquitoes (0.031 ppm) and the labeled rate (1.6 ppm). Susceptibility to spinosad did not differ significantly between

the susceptible and field-collected mosquitoes; however, there were marked differences among immature stages of the non-target taxa. Susceptibility was greatest in the mayfly, followed by the damselfly, then the dragonfly.

205 Field evaluation of Natular™ XRT in catch basins in Chicago, Illinois
Justin Harbison, jharbison@luc.edu

Over the past 4 yr the North Shore Mosquito Abatement District (NSMAD) has performed field trials on spinosad-based Natular™ XRT catch basin tablets. This formulation is marketed to be effective for up to nearly 26 wk (180 days) by slowly releasing the AI over time. Generally the NSMAD found the duration of effective control from XRT tablets to be much shorter than the expected 26 wk. Additionally, observations from approximately 200 treated basins in the 2014 trials suggest that during a particularly rainy season a significant portion of basins (over 70%) lose their larvicide treatments 1 to 17 wk after application through rapid dissolution or flushing.

206 Field evaluation of Natular products in Merced County, California
Jason Bakken, mcmadbiologist@unwiredbb.com

In a blind study in 2008, Merced County MAD worked with 2 formulations of what were later disclosed as Natular EC and Natular G. These formulations were hand applied against *Aedes nigromaculis* larvae in irrigated pastures and *Culex tarsalis* in seasonal wetlands. The EC provided 100% control of *Ae. nigromaculis*, 48 h after treatment. The G resulted in 99% larval reductions of *Ae. nigromaculis*, 72 h post-treatment. As for controlling *Cx. tarsalis* larvae, the G provided 98% control in wetland plots, while the EC provided variable results. The following season Natular G (0.5% granule) and Natular 2EC (20.62%) were aerially applied to irrigated pastures. Natular G reduced larval populations by 99%, 48 h after treatment, while Natular 2EC produced mixed results. In 2010, the Merced County MAD introduced Natular 2EC into the aerial larviciding regimen. Control of *Ae. nigromaculis* larvae in irrigated pastures and *Ae. melanimon* in recently flooded wetlands was variable. A summary of 4-yr of operational use of Natular products will be presented.

207 Operational use of spinosad products for mosquito control in Volusia County, Florida
Suzanne Bartlett, sbartlett@volusia.org

Volusia County Mosquito Control (VCMC) in central Florida utilizes an integrated mosquito management (IMM) program, not unlike many programs across the US. Larviciding is a vital component of IMM in Volusia County and takes place across a wide variety of habitats including but not limited to tidal wetlands, freshwater swamps, artificial containers, and catch basins. To ensure the efficacy of the active ingredients associated with larviciding in Volusia County, the district utilizes a rotation of actives that includes *Bacillus* formulations, methoprene and spinosad. Four (4) spinosad formulations are currently utilized and include tablet, granular and liquid formulations. Both the liquid (Natular 2EC) and granular formulations (Natular G and G30) are applied aerially via helicopter and from ground-based equipment, including a Buffalo Turbine. The G30 formulation is used as a pre-treatment in many areas of the district that produce *Aedes taeniorhynchus, Psorophora columbiae* and *Culex nigripalpus*. Containers in "super production" sites such as cemeteries and salvage yards are routinely treated with G30, with efficacy exceeding label specifications. The presentation will discuss the value of spinosad within VCMC's IMM program to resistance management and the efficiency of operations.

208 Laboratory and semi-field evaluation on spinosad for mosquito control in California
Mir Mulla, Mir.mulla@ucr.edu

Although the mosquito larvicidal activity of spinosad was identified back 30 years ago during the early-stage insecticide screen against *Aedes aegypti*, its potential use for controlling mosquitoes was not investigated until the last 10 years. Early 2005, our laboratory initiated a study to determine the susceptibility of *Aedes* and *Culex* species to technical material and a liquid concentrate formulation (120 SC) of spinosad in the laboratory and later on to evaluate the efficacy of the formulation in semi-field conditions (microcosms and mesocosms) against *Culex* species, in collaboration with Dow AgroSciences. In the laboratory, spinosad technical powder was highly active against 2nd and 4th instars of *Ae. aegypti* and *Culex quinquefasciatus* after 24 h of exposure. The extent of mortality increased slightly after 48 h of exposure. Second instars were slightly more susceptible than 4th instars. The liquid formulation showed somewhat higher activity (about 2X) than the technical powder material at both the LC_{50} and LC_{90} levels. In the field microcosm tests against natural populations of mosquitoes, the liquid formulation yielded excellent control of immature *Culex* spp. for 21 days at concentrations of 0.05 mg AI/liter and 35 days at 0.1 to 0.5 mg AI/liter in outdoor tubs. This formulation also yielded excellent control of natural *Culex* mosquitoes for 14 days or longer at 0.025 to 0.1 mg AI/liter in outdoor ponds. Our lab and field data demonstrated that spinosad has a good potential to be a new class of mosquito larvicide and has proved so.

Push-Pull-Killing Strategy for Adult Mosquito Control II

209 Field efficacy of an autodissemination station against *Aedes albopictus*
Devi Suman, dssuman37@gmail.com, Isik Unlu, Eric Williges, Gregory Williams and Randy Gaugler

The Asian tiger mosquito, *Aedes albopictus,* is a container mosquito transmitting chikungunya and dengue viruses. This mosquito prefers to oviposit in multiple small to medium-sized cryptic habitats in urban environments. This behavior creates hurdles for conventional chemical control strategies, which often have low penetration to these habitats. We have developed an autodissemination station (ADS) that attracts gravid females (pull), contaminates them with high concentration AI (20% on oil band and 60% on powder band) of insect growth regulator (IGR: pyriproxyfen), and allows them to exit and transfer the IGR to their larval oviposition site (push). The station efficacy was tested under different field conditions such as hot-spot, cryptic habitat and backyard using natural mosquito populations during the mosquito season of 2013-14. Stations were constructed with self-maintaining features such as water auto-refilling and reservoir of oil formulation. The stations were deployed in the field with sentinel cups and the water from sentinel cups was sampled on 1, 2, 4 and 8 wk post-deployment of stations for larval bioassay. Efficacy of the station was measured as pupal mortality by IGR-bioassay of field samples using 3rd instar *Ae. albopictus* under standard laboratory conditions. Our results showed various degrees of pupal mortality and the pyriproxyfen transfer was confirmed by residue analysis. Our data also indicated successful transfer of the IGR to the cryptic habitat where traditional pesticide spray had poor penetration. The technique may be used as a part of the IPM program against Asian tiger mosquito as well as other container mosquitoes such as *Ae. aegypti*.

210 Use of attractants and traps in attract (pull) and kill strategies
Daniel Kline, dan.kline@ars.usda.gov

Recently we have invested much effort in the development and/or evaluation of new attractant and trapping technologies for the management of mosquitoes and other biting flies (e.g., biting midges and tabanids). Various attractants, trap types and insecticide-treated targets were evaluated. We have discovered that the concept, "one size fits all," does not apply for any group of target insects in regard to trap, target or attractant combination. Some examples of effective trap/target-attractant combinations will be presented for each group of target insects.

211 Traps used for pulling/killing adult mosquitoes in Anastasia Mosquito Control District
Mike Smith, mksmithamcd@bellsouth.net and Rui-De Xue

Mosquito magnets, New Jersey light traps, Mos Hole traps, and BG traps have been used and evaluated for pulling and killing adult mosquitoes and midges by Anastasia Mosquito Control District, St. Johns County, FL in certain areas (recreational parks, school, residential subdivisions) and special events. The trap methods and techniques benefited the environments and reduced populations of nuisance and vector mosquitoes and pestiferous midges, and was appreciated by local residents.

212 Evaluation of new active ingredients for use in attractive toxic sugar baits for mosquito control
Gunter Muller, guntercmuller@hotmail.com and John Beier

Laboratory tests were conducted to determine the efficacy of new inert active ingredients against *Culex pipiens* and *Aedes aegypti* to further increase the arsenal of active ingredients that can be used in attractive toxic sugar bait (ATSB) methods. Additional laboratory experiments were conducted to evaluate the ability of mosquitoes to penetrate a film biolayer for incorporation into bait station design. The biolayer will prevent contamination of the attractive toxic mixture contained in the bait station. Overall, 3 new inert active ingredients were identified in successfully knocking down greater than 90% of both *Culex pipiens* and *Aedes aegypti* populations within 48 h. Significant mortality was recorded in *Cx. pipiens* populations following the presentation of an ATSB bait station (0.02% dinotefuran AI) covered with the biofilm. This suggests that the mosquitoes can successfully penetrate the biofilm and imbibe the bait.

213 Attractive toxic sugar baits mixed with attractants to increase pull/kill efficacy against *Aedes* mosquitoes
Rui-De Xue, xueamcd@gmail.com, Jodi Scott, Ali Fulcher, Sandy Allan and Dan Kline

Attractive toxic sugar baits (ATSB), active ingredient of 1% boric acid, mixed with mosquito attractants (octenol and lactic acid) were evaluated in the laboratory and semi-field against *Aedes aegypti and Ae. albopictus*. The mixtures of ATSB and attractants increased number of mosquitoes tested by the olfactometer, compared with the original ATSB; also, the mixtures increased mortality of adult mosquitoes, but the species showed significant impact on the attraction and mortality. This preliminary study showed some prospects to improve ATSB's attraction and mortality, and may reduce non-target impact.

213a Evaluation of the effects of an agricultural spreader/sticker included in attractive toxic sugar bait on mosquito and non-target arthropod populations
Jodi Scott, bowerj@ufl.edu

Attractive toxic sugar baits (ATSB) are a novel way of greatly reducing mosquito populations in an area. Many of the studies concerning ATSB methods have shown a wide range in duration of the field application efficacy. The ranges have been from 21 days to upwards of 4 wk depending on the species. The range in efficacy duration may be attributed to the vast geographical differences and climate of different areas of application. To circumvent weather issues, such as rain, an agricultural spreader sticker (Poly Control-2) was integrated, at the lowest label rate, into an ASB/ATSB. Before testing the efficacy of controlling mosquito populations, the Poly Control-2 was added to attractive sugar bait (ASB) and evaluated for possible risks to non-target insects under field conditions. Less than 3% of the overall non-target arthropods in the area ingested the bait. Directly following the non-target studies, Poly Control-2 was added to ATSB (1% boric acid) and applied to the same 150 m site as the non-target study. A control site (150 m), 66 m from the ATSB site, was sprayed with ASB. Surveillance was conducted with 3 CDC light traps in each site, collected twice a week for 2 wk prior to application, and twice a week for 4 wk post-application. ATSB application controlled 59% of the mosquito population to 4 wk post-application. *Psorophora columbiae*, *Aedes infirmatus*, and *Coquillettidia perturbans* had ~ 75% control of their mosquito populations in treatment sites 4 wk post-application.

Recent Developments in Mosquito Surveillance and Trapping Techniques II

214 The gravid *Aedes* trap: application of a "crab pot" design for no-fuss collection of gravid *Aedes*
Scott Ritchie, scott.ritchie@jcu.edu.au, Alvaro Eiras and Laila Heringer

We recently developed novel traps that capture large numbers of mosquitoes without the use of power. These rely upon luring mosquitoes into a translucent "passive" trap from which those mosquitoes, attracted to the light entering the translucent body, cannot readily find their way out. A crab pot for mosquitoes so to speak. Passive box traps baited with CO_2 have proven successful in capturing large numbers of *Aedes, Anopheles, Culex* and *Verrallina* mosquitoes. But current surveillance methods for container-breeding *Aedes* such as *Ae. aegypti* and *Ae. albopictus* rely upon powered traps (BG Sentinel, aspirators) or adhesives (sticky ovitraps). No one likes messy glue. So, to replace the sticky ovitrap, we adopted the passive trap approach to create a glue-free gravid trap. The gravid *Aedes* trap (GAT) employs a black bucket containing infusion to attract and lure gravid *Aedes* into a translucent top. Like an insect in a room that flies to a window, the mosquitoes are attracted to the light from the walls of the translucent top. We treat the inside of the top with insecticide as a surface spray or insecticide treated bednet to knock down the mosquitoes. In field trials, the GAT captured significantly more gravid *Ae. aegypti* than the sticky ovitraps we tested it against. We have shown that the trap captures gravid *Ae. albopictus* and *Ae. polynesiensis.* Mosquitoes captured by the GAT can also be processed for dengue virus and *Wolbachia* endosymbiotic bacteria.

215 The Suna trap – an odor-baited tool for mosquito monitoring and control
Alexandra Hiscox, alexandra.hiscox@wur.nl, Niels Verhulst, Bruno Otieno, Anthony Kibet, Philemon Omusula, Collins Mweresa, Wolfgang Mukabana and Willem Takken

The development of synthetic odor baits that are as attractive to the African malaria vector (*Anopheles gambiae*) as natural human odor has created an opportunity for the development of new odor-baited mosquito traps. These traps function as standardized tools for mosquito monitoring, but also have the potential to be used as tools for malaria vector control. The development of the Suna trap is described from the initial concept through the processes of laboratory, semi-field, and field testing, finally culminating in the wide-scale introduction of solar-powered traps for malaria control on Rusinga Island, western Kenya.

Under semi-field conditions, the Suna trap performed as well as the CDC light trap when used to capture mosquitoes inside an experimental house. When used outside houses in the field, Suna trap catch sizes of *An. funestus* and *An. arabiensis* equalled those of an odor-baited MM-X trap. Subsequent development of the trap under laboratory conditions led to the creation of a mosquito-holding chamber where trapped mosquitoes can be held alive for at least 24 h with low levels of mortality, an important advancement in the use of Suna traps to monitor arboviral vectors.

The use of Suna traps in rural regions of East Africa, Southeast Asia and South America provides opportunities for further improvement of odor blends, as well as the evaluation of trap performance against different mosquito species.

216 Platform MI-Dengue: 10 years of experience of citywide surveillance of adult *Aedes aegypti* in Brazil

Alvaro Eiras, Alvaro.Eiras@gmail.com

Monitoring adult *Aedes aegypti* abundance with fixed position traps has been considered as an alternative surveillance method that shows promise for directing vector control and predicting when and where dengue outbreaks will take place. In partnership between the university and a "spin-off" company, we developed the MI-Dengue Platform. The platform consists of an integration of continuous vector monitoring, at fine spatial and temporal scales, and information technology platform for near real-time data collection, analysis, and decision-making. The surveillance data generated from the system were used to calculate weekly vector indices and easily discernible geographical hot spots to target vector control resources. Once identified, directing control efforts to high priority areas with virus detection may reduce future virus circulation. Citywide analysis also facilitates the detection of novel serotype introduction to the city, which can be an indicator for dengue epidemics. Furthermore, the viral analysis of mosquitoes is low-cost and can be implemented with a simple laboratory protocol. Results from 21 cities in Minas Gerais State, Brazil, indicated that over 27,000 dengue cases were prevented (2.7 times lower than conventional methods) and about US $7 million were saved in direct and indirect costs. MI-Dengue Platform is an alternative for monitoring other urban vector diseases such as chikungunya. The 10 yr experience of MI-Dengue in Brazil covered over 50 Brazilian cities and in 2014, a pilot test was conducted in Lee County, FL (USA).

217 Surveillance for chikungunya virus vectors in New Orleans, Louisiana

Sarah Michaels, srmichaels@nola.gov, Brendan Carter, Laura Turpin, Cynthia Harrison, Mieu Nyugen, Kevin Caillouet, Justin Davis and Claudia Riegel

New Orleans, LA has abundant populations of the chikungunya virus (CHIKV) vectors, *Aedes aegypti* and *Ae. albopictus*. In the summer of 2014, CHIKV was diagnosed in 3 individuals with travel history to the Caribbean; in 1 of these cases the individual was viremic. Vector surveillance was initiated using aspirators and BG Sentinel® traps (BGS) in close proximity to travel-related CHIKV cases and in areas of the city known to have high vector mosquito populations. Mosquitoes were tested for CHIKV by RT-PCR and collection data was evaluated for ground and aerial adulticide application efficacy. No mosquito pools tested positive for CHIKV. Collections after aerial adulticiding operations were significantly lower than collections made prior in some areas, while others had little or no observed effect. These results were likely affected by neighborhood and vegetation density, urban obstructions and baseline mosquito productivity. The continued threat of CHIKV introduction remains, and our experience suggests that it is important for municipalities to identify and monitor local *Aedes* populations through routine surveillance activities using the most appropriate tools available.

218 Incorporating Biogents BG Sentinel traps into East Baton Rouge Parish Mosquito Abatement and Rodent Control District's adult mosquito surveillance program

Randy Vaeth, rvaeth@brgov.com, Michael Morganti and Todd Walker

Our abatement program in East Baton Rouge Parish, LA has employed a variety of collection techniques over the years in an effort to sample *Aedes albopictus* adult populations. Aspirator collections have proven to be labor intensive; ovitrap sampling frequently is inconsistent for us when competing oviposition sites are present; Fay-Prince traps are bulky, and we have had limited success with modified gravid traps since we use fish oil emulsions to attract *Culex quinquefasciatus*. The reported widespread successes of the BG-Sentinel Trap and lure for collecting *Ae. albopictus*, encouraged us to participate in 2 evaluation studies during the 2014 season. We conducted 16-day trials comparing the BG-Sentinel 1.0 to the BG-Sentinel 2.0 using either CO_2 or no attractant. We also evaluated the BG-Sentinel 2.0 traps and the BG lure with different trap outer wrappings for 12 days at 2 different sites. The CO_2 baited BG-Sentinel 1.0 did collect significantly more ($p<0.05$) *Ae. albopictus* than the similarly baited BG-Sentinel 2.0. We found no significant differences in *Ae. albopictus* collections between the BG-Sentinel 2.0 traps with either black or white trap outer wrappings. In the future, we hope by employing these traps that we will enhance our efficiency in collecting this important nuisance and disease vector species, not so much for disease surveillance, but to help estimate the efficacy of our control intervention methods.

219 Comparison of Biogents® BG Sentinel 2.0 and BG Sentinel 1.0 in trapping mosquitoes with 3-component cartridge lure and BG mesh lure

Hanayo Arimoto, Hanayo.Arimoto@med.navy.mil, Alec Richardson and Peter Obenauer

The trapping efficacy of Biogents® BG-Sentinel 2.0 (BG2) and BG-Sentinel 1.0 (BG1) in sampling wild mosquito populations was compared. Both types of traps were paired with BG 3-component cartridge lure and BG mesh lure. All traps were baited with CO_2 in the form of dry ice. This study was conducted in a yard in an urban neighborhood in Jacksonville, FL, USA. All 4 trap combinations and 2 control traps were spaced 20 m apart and rotated daily for 18 trapping days. Specimens were collected every 24 h. Overall, a total of 4,435 mosquitoes were trapped and identified to 19 different species. At 56.1%, *Culex nigripalpus* was the

predominant species of mosquitoes caught. *Aedes albopictus* comprised 10.1% of the total trapped. BG1 with 3CL caught the majority of the *Ae. albopictus* (28.9%). The comparability and efficiency of BG2 with 3CL and BG1 with BGM are discussed in greater detail.

The views expressed in this abstract are those of the author and do not necessarily reflect the official policy or position of the Department of the Navy, Department of Defense, nor the U S Government.

Spinosad for Mosquito Control: From the Laboratory to the Field II

220 Laboratory study of the influence of water temperature on spinosad efficacy against *Culex pipiens* larvae

Nayer Zahiri, nzahiri@smcmad.org

Spinosad, a naturally occurring product of the fermentation of the bacterium *Saccharopolyspora spinosa*, contains a mix of 2 spinosoids: spinosyn A, the major component, and spinosyn D the minor component, in an approximately 17:3 ratio. Spinosad is a highly effective bioinsecticide by both contact and ingestion to numerous insect species. Recently some formulations have been developed to control mosquito and midge larvae. In this laboratory study, the influence of water temperature ranging from 15 to 35°C on the activity and efficacy of various formulations of spinosad control agents was studied against laboratory reared mosquitoes larvae of *Culex pipiens*. The activities of spinosad granule, spinosad tablets and spinosad XRT formulations against 3rd and 4th instar larvae were markedly affected by water temperature, indicating that at cooler water temperatures the level of efficacy was lower than at high temperatures. The efficacy of spinosad-based products was temperature-dependent and based on this preliminary study, spinosad products should be used at moderate to high temperatures.

221 Evaluation of spinosad products in urban and rural field environments

Jennifer Henke, jhenke@cvmvcd.org and Gregory White

Application of mosquito control products in the field is rarely performed under ideal conditions, and determining how those conditions impact product efficacy is important in integrated vector management. We examined the use of spinosad tablets with expected long-residual activity (Natular™ XRT) in urban catch basins that received daily inputs of irrigation run-off. Follow-up visits examined the mosquito and non-target populations as well as water parameters of temperature, pH, conductivity, and dissolved oxygen. The residual efficacy was typically 90 days, about half of the potential 180 days on the label. We believe the cause of the shortened length of activity was the continual input of water into the catch basins. We also examined field applications of a spinosad-based, multi-brood granular formulation with Natular™ G30. These applications were made using a helicopter in rural duck clubs and wetlands in early March to help reduce the springtime peak of mosquitoes. Efficacy of the larvicide application was evaluated by monitoring the number of larval and adult mosquitoes following the treatment. The results indicated that the use of Natular™ G30 product appeared to have reduced the number of vectors in this cool period early in the mosquito season.

222 Field evaluation on Natular products in subtropical China: a collaborative effort with WHOPES

Qiyong Liu, liuqiyong@icdc.cn

Spinosad is a macrolide biopesticide from the fermentation liquid of *Saccharopolyspora spinosa*. However, very little is known regarding the effect of Natular products on the vector mosquitoes in China. The objective of this study was to evaluate efficacy of Natular G30 and T30 in small- and large-scale trials to determine the optimal field application rate as well as initial and residual efficacy against *Culex quinquefasciatus* in natural polluted aquatic habitats, and against *Cx. tritaeniorhynchus* and *Anopheles sinensis* in natural or/and manmade aquatic settings. The present study was conducted in Haikou, Hainan, China. The mean number of pupae and larvae per dip from each type of habitat was calculated for each sampling day for each treatment as well as control. The 1st and 2nd instars were grouped into young instar larvae and the 3rd and 4th instars into old instar larvae. Percentage reduction of young and old instar larvae and pupae were estimated using Mulla's formula. Physico-chemical and weather parameters were recorded during the period of this study. We found that Natular T30 has long persistence against mosquito larvae in containers such as glass, ceramics, and cement pools. Natular G30 has better mosquito control in paddy field, because sediments have a serious adverse impact on the efficacy of Natular T30. The quality of water has the similar effect on Natular G30 and T30. There was no negative effect of Natular G30 and T30 on aquatic non-targets (fish, tadpoles and frogs) under the recommended concentrations. Natular G30 and T30 also killed chironomid larvae under the recommended concentrations for mosquito control. The results indicate that Natular T30 and G30 are significant tools for the control of mosquitoes and mosquito-borne diseases in the world.

223 Spinosad: efficacy and persistence against container inhabiting mosquitoes
Scott Crans, scott.crans@rutgers.edu, Isik Unlu, Taryn Crepeau, Gregory Williams, Ary Faraji, Healy Sean, Lisa Reed and Randy Gaugler

Determining inhibition of larval development of invasive container-inhabiting mosquitoes of public health importance was a primary goal of this study. For this, plastic containers commonly found in the back yard residential setting were treated with spinosad (Natular G30) covering the low, middle and maximum labeled range and compared to an untreated water control. Persistence of insecticidal activity was evaluated with respect to relative sunlight exposure, water temperature, water pH, accumulation of organic debris, and container effects. Ten species of container inhabiting mosquitoes were collected in our simulated larval habitats. The 4 most abundant species accounting for 81% of the mosquitoes sampled were invasive exotics, *Aedes albopictus* and *Ae. japonicus,* followed by the West Nile virus vectors *Culex pipiens pipiens* and *Cx. restuans.* Spinosad treatment was extremely effective at inhibiting development of container inhabiting mosquitoes at all application rates tested with persistence (>95% control) lasting beyond the 30 day extended release formulation under all study conditions tested. The interactions between habitat water pH, organic loading and sunlight exposure with respect to spinosad persistence varied on a site specific basis. There were no significant container induced effects observed with respect to *Ae. albopictus* development through pupation in the laboratory. Dominant cover vegetation, exposure to direct sunlight and larval habitat water pH are factors to be considered on a site-specific basis with respect to spinosad application rates. This extended release spinosad formulation warrants consideration as a rotational public health pesticide for integrated mosquito management programs.

224 Profile of resistance and cross resistance in mosquitoes to spinosad
Tianyun (Steven) Su, tsu@wvmvcd.org and Min-Lee Cheng

A *Culex quinquefasciatus* Say colony was established from surviving late instars and pupae from a semi-field evaluation on Natular® XRG (2.5% spinosad). Selection pressure was applied at LC_{70-90} levels to 10,000-15,000 of late 3rd and early 4th instar larvae each generation with Natular XRG. At F60, Resistance Ratio (RR) reached 14,363 – 16,622 fold at LC_{50} and 128,872 – 140,065 fold at LC_{90} in response to continuous selection. The spinosad-resistant *Cx. quinquefasciatus* was found not to be cross-resistant to *Bti*, a combination of *Bti* and *B. sphaericus*, methoprene, pyriproxyfen, diflubenzuron, novaluron, temephos, imidacloprid or indoxacarb. However, it showed various levels of cross-resistance to *B. sphaericus*, spinetoram, abamectin and fipronil. *Culex quinquefasciatus* highly resistant to *B. sphaericus* or moderately resistant to methoprene did not show cross-resistance to spinosad and spinetoram. The information generated from this study is immediately applicable to mosquito control operations.

Poster Session Abstracts

Adult Control

P-01 Duration of effectiveness of permethrin-treated clothing for prevention of mosquito bites
Stephanie Richards, richardss@ecu.edu, Jo Anne Balanay, Jonathan Harris, Victoria Banks and Steven Meshnick

This study followed up on a previous field study showing a reduction of effectiveness of permethrin-treated clothing after 1 year of wear and we evaluated the extent to which environmental conditions impact mosquito knockdown for permethrin-treated fabrics. We evaluated the extent to which fabric type (100% cotton denim jeans, 100% polyester work shirt, 35% cotton/65% polyester United States Forester uniform work shirt), light exposure (0 or 100%), temperature (18°C, 32°C), and number of washes (0, 3, 12, 36) affected mosquito knockdown 2 h post-exposure (hpe), mosquito mortality 24 hpe, and permethrin content. Washing and fabric type significantly affected permethrin content. Light alone did not affect permethrin content; however, permethrin content was impacted when fabrics were subjected to washing treatments under different lighting conditions. Denim fabric having no washes and no light exposure showed the highest amount of permethrin. Washing and light exposure significantly reduced the ability of permethrin-treated fabric to induce mosquito knockdown and/or mortality. Temperatures tested showed no effects on permethrin content or mosquito knockdown and mortality. Long-lasting impregnation of uniforms provides effective protection against biological hazards such as mosquito bites, albeit for less than 1 yr. The risks of mosquito and tick borne disease is high among outdoor workers, such as foresters, throughout the world. Employers and employees should recognize occupational health risks and consider the use of permethrin-impregnated clothing and uniforms.

P-02 The use of CDC autocidal gravid oviposition traps (CDCAGO) to control *Aedes aegypti* in an urban residential community in Clovis, CA

Jodi Holeman, jholeman@mosquitobuzz.net, Steve Mulligan, Charlie Smith and Anthony Cornel

In 2013 the Consolidated Mosquito Abatement District (District) utilized the novel CDC Autocidal Gravid Oviposition trap (CDCAGO) to monitor abundance and dispersal and found the trap to be highly effective for both aims. The use of CDCAGOs as a control method to suppress *Aedes aegypti* populations was demonstrated to have measurable success in San Juan, Puerto Rico (Barrera 2014). In 2014, we tested the effectiveness of these traps at a reduced density to control a recently established population of *Ae. aegypti* in an urban residential community in the City of Clovis, Fresno County, CA. Overall changes in the adult *Ae. aegypti* population were monitored by BG Sentinel traps (Biogents, Regensburg, Germany) situated within the targeted intervention area as well as areas with similar landscape and abundance outside of the intervention area. Although homeowner compliance with CDCAGO placement at all 146 properties within the study area was achieved, there was no significant difference in the average number of *Ae. aegypti* collected in BG Sentinel traps between the intervention and non-intervention areas. However, there was a significant reduction in the number of adults collected in CDCAGOs in the final 2 wk of the study when compared to the previous 4. This reduction in CDCAGO collections observed over time suggests that the placement of CDCAGOs at greater densities could reduce the number of older gravid females in the population and lower the potential for disease transmission.

P-03 Chemical composition, adulticidal and repellent activity of essential oils from *Mentha longifolia* L. and *Lavandula dentata* L. against *Culex pipiens* L.

Ali al-sarar, asarar@ksu.edu.sa

The essential oils from *Mentha longifolia* L., and *Lavandula dentata* L. were evaluated for their insecticidal and repellent activity against adult females of *Culex pipiens* L. The 2 oils showed LC_{50} values of 0.215 and 0.217 µl/l, respectively. *Lavandula dentata* oil showed higher repellent activity than *Mentha longifolia* oil against adults of *Cx. pipiens*; paraffin oil significantly prolonged the protection time for the 2 oils. The longest protection time was 165 min for *L. dentata* oil, at 1 µl/cm2, when the tested oils were applied in paraffin oil. The principle compounds in the 2 oils were pulegone, l-menthone, 1-8-cineole, eucarvone and borneol for (*M. longifolia*); and camphor, fenchone, linalool and fenchol for (*L. dentata*). The 2 oils were effective as repellent substances when compared with commercial materials. Further studies are needed to isolate the most effective mosquito control agents from these oils.

P-04 Novel passive device for volatile insect attractant and repellent release

Jedidiah Kline, jdkl81@ufl.edu, Bradley Willenberg, Phillip Koehler, Daniel Kline, Joyce Urban and Christopher Batich

At the University of Florida Materials Science and Engineering Department, we have developed a completely non-powered device to release chemicals for attracting and repelling mosquitoes. The technology relies on a 2 reservoir design. The inner reservoir stores a range of active ingredients, and is impermeable to chemicals commonly used as spatial repellents or insecticides. The device is composed of cotton, aluminized polyethylene, and the desired active ingredient. An inner reservoir stores the active ingredient until ruptured by the researcher, while an outer reservoir and wicks control the release rate. Until that time, the device can be handled without exposure to the insecticide or repellent.

Unlike currently available devices such as the Terminix Allclear device or SC Johnson's OFF! Clip-On, which rely on fans and batteries, and other products such as the Therma-Cell device which relies on heat generated by butane gas, our device is completely non-powered. It is rugged and ideal for field conditions. This, combined with our dual reservoir technology, makes our device unique to the best of our knowledge. We believe it will be of great interest to mosquito control and public health professionals for surveillance, repellent, and attractant applications.

P-05 Investigating the effect of droplet size on deposition on caged mosquitoes and their mortality

Jennifer Wright, jennifer.wright@med.navy.mil and James Cilek

A study was conducted to investigate the effect of sprays of various volume medium diameters (VMD) on pesticide efficacy by deposition on adult mosquitoes and mortality. Anvil 10 + 10 diluted 1:375 with corn oil was sprayed to caged female *Aedes aegypti* using sprays with droplet spectra (VMDs) of 8, 10, 12, 16, 20, 25, 30, 35, and 40 µm at an application rate of 0.415 oz/acre selected for <100% mortality. These sprays were repeated with corn oil as control and a mixture of corn oil and Uvitex OB fluorescent dye for deposition on mosquitoes. For each application, 1.5 ml of liquid was sprayed which was replicated 3 times. Mortality for all applications was determined at 4 and 24 h after treatment. For deposition, the mosquitoes in each cage were killed by freezing, counted, and the amount of oil deposited/mosquito determined by fluorometry.

Mosquito mortality from the corn oil spray showed increase with droplet size from 8 to 12 µm VMD then decreased and leveled after 30 µm VMD. The mosquito mortality from the Anvil 10 + 10 application was

corrected for mortality from the control for each droplet spectrum. Overall, there was no effect of droplet size on mosquito mortality and no clear trend was found. However, deposition of corn oil on mosquitoes was significantly affected by droplet size. The lowest deposition resulted from 10 µm VMD spray while the highest resulted from 12 µm VMD. The general trend of the data showed that deposition increased with increase in VMD up to 12 µm and then decreased which is consistent with mosquito mortality due to the corn oil only spray.

Disclaimer: The views expressed in this presentation are those of the author and do not necessarily reflect the official policy or position of the Department of the Navy, DoD, nor the U. S. Government.

P-06 Use of multiple methods of resistance monitoring of urban *Culex quinquefasciatus* populations show significant levels of resistance at phenotype and genotype to pyrethroids
Gregory White, gwhite@cvmvcd.org and Melissa Snelling

In order to ensure public health pesticides are effective against target vector populations, resistance levels were investigated in urban populations of *Culex quinquefasciatus* mosquitoes. Populations of mosquitoes were tested for resistance to 3 adulticide products: Anvil 10+10, Scourge 4+12, and Pyrocide 7396. Mosquitoes were tested for resistance in bottle bioassays and then using ULV applications with mosquitoes in sentinel cages. The sentinel cage ULV trial showed an operationally significant result to the pesticide showing the highest levels of resistance in the bottle bioassay. The mosquitoes were then sent for *kdr* testing at the California Department of Public Health Laboratory. Genetic testing showed the population with the highest levels of resistance in the bioassays had the highest percentage of the *kdr* mutation. These populations of *Cx. quinquefasciatus* mosquitoes had only been treated with adulticide applications on a few occasions since 2003, when West Nile virus was found in urban areas of the Coachella Valley. The infrequent adulticide treatments by the District indicate that the selective pressure to pyrethroids is coming from another source.

Behavior/Biology

P-07 Do capture data from mosquito traps represent reality?
Donald Barnard, don.barnard@ars.usda.gov

Collectively, the effects of mechanical trap style, the method of trap placement in the field, mosquito activity phase, and other biological phenomena are manifest as sample bias that leads to vector detection failure(s) and/or erroneous predictions of mosquito activity. The goal of this research is a practical (science-based) system for the selection of mosquito trap type and the (geo-coordinate-based) placement of mechanical traps in the field when such traps are used for adult mosquito detection and surveillance. Two objectives have been undertaken to achieve this goal. The first objective is to determine the species sensitivity and sample bias characteristics of different styles of mechanical traps. The second objective is to design and test strategies for the placement of mechanical traps in the field. The principal criterion for success in this endeavor is the accuracy of spatio-temporal predictions of vector activity based on the species diversity and adult mosquito density data obtained from mechanical traps.

P-08 Effect of botanical components of 18 plant species on *Aedes aegypti*
William Dees, wdees@mcneese.edu, Janie Theriot, Caleb Ardizzone, Allison Fusilier, Omar Christian, Cecilia Richmond, Jill Hightower and Janet Woolman

The Medical Entomology Research Laboratory at McNeese State University is investigating if components and/or derivatives (e.g., extracts) of plants native to Louisiana alter the behavior and development of medically important arthropods (e.g., mosquitoes and nuisance flies). Information obtained from these investigations may lead to innovative area-wide pest management methodologies as well as novel personal protective measures against biting arthropods. Current studies focus on the effects of botanical components on mosquito mortality. We evaluated the effects of freshly-cut plant parts from 8 plant families on female *Aedes aegypti* mosquitoes. Plant families included Apiaceae, Apocynaceae, Asteraceae, Euphorbiaceae, Lamiaceae, Lythraceae, Malvaceae, and Verbenaceae. Standard plastic Petri dishes were used to hold mosquitoes and cut plant parts from 18 plant species. We recorded percent mortality at 24 and 48 h. Mosquitoes exposed to fresh-cut flowers/petals, buds, leaves, stems, and seeds from Apiaceae, Asteraceae and Lamiaceae exhibited over 50% mortality when compared with the controls. Genera of interest include: *Chrysanthemum*, *Eryngium*, *Eupatorium*, *Rudbeckia*, *Monarda*, and *Pycnanthemum*. Mosquitoes exposed to different parts of a chrysanthemum plant (flowers, buds, leaves, and stems) exhibited 100% mortality in 24 h. Mosquitoes exposed to cut buds of *Pycnanthemum muticum*, *P. tenuifolium*, and *Monarda fistulosa* exhibited 100% mortality in 24 h.

P-09 Oviposition response of *Aedes aegypti* to Louisiana native plant extracts

William Dees, wdees@mcneese.edu, Jesse Dupre, Adam Richard, Cecilia Richmond, Omar Christian, Caleb Ardizzone, Jill Hightower and Janet Woolman

We evaluated the effect of Louisiana native plant extracts on ovipositing *Aedes aegypti* mosquitoes. Bioassays were conducted in screened cages (46 X 46 X 46 cm) containing plastic cups lined with germination paper on which eggs were deposited. Each cup contained 60 mL distilled water and 10 µL of hexane, acetone or methanolic plant extracts. Extracts were made from the following plant parts: (1) stems and roots from Cluster Bushmint, *Hyptis alata*; (2) stems and roots from Wooly Rose Mallow, *Hibiscus lasiocarpus*; and (3) buds, stems and roots from Rattlesnake Master, *Eryngium yuccifolium*. The bioassay was conducted for 24 h. After 24 h, the germination paper in each cup was removed and the eggs on each paper were counted. We calculated the oviposition activity index (OAI) for each extract. Hexane extracts of *E. yuccifolium* buds and roots and the acetone extract of *E. yuccifolium* stems repelled ovipositing mosquitoes. Ovipositing mosquitoes were slightly attracted to hexane extracts of *E. yuccifolium* stems, methanolic extracts of *Hyptis alata* roots and methanolic extracts of *E. yuccifolium* stems and roots. Ovipositing mosquitoes demonstrated the greatest attraction to acetone, hexane, and methanolic *Hibiscus lasiocarpus* root extracts. The OAI ranges for the *H. lasiocarpus* root extracts were (+)0.32-0.41 on a scale of (+)1 (attraction) to (-)1 (repellency).

P-10 Seasonal variation in body size of *Aedes vexans* and *Culex tarsalis* collected in CO_2 baited CDC miniature light traps in northeastern Colorado

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Adult mosquito size, as estimated by wing length, is influenced by many environmental factors including larval density, nutrition and rearing temperature. Smaller size has been shown to negatively affect survival, vectorial capacity and host seeking behavior. In Colorado, *Aedes vexans* utilize a range of larval habitats with temporary shallow pools predominating while *Culex tarsalis* often use relatively stable cattail marshes and other wetlands. Because *Cx. tarsalis* oviposit in more stable and homogeneous sites than *Ae. vexans*, we hypothesized that the latter species would exhibit higher variation in size over the climatologically dynamic season. *Culex tarsalis* and *Ae. vexans* did not differ in seasonal variance of body size. However, simple regressions between wing length and physical factors experienced during their larval development 3-11 days previously indicated that body size was negatively correlated with total precipitation in *Ae. vexans* and negatively correlated with mean ambient temperature in *Cx. tarsalis*. Our preliminary results indicate that post-hatch precipitation and temperature during larval development are important variables in the development of adult body size.

Disease/Vector Studies

P-11 Mosquitoes collected in a rural area of central Mississippi and implications for WNV transmission

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To determine abundance and seasonality of potential WNV mosquito vectors in a forested area of central Mississippi, mosquitoes were collected weekly from a wildlife management area located approximately 10 mi from a local urban area with numerous human WNV cases. We were particularly interested in the presence or absence of *Culex quinquefasciatus*, the primary vector of WNV in Mississippi, although other *Culex* species were assayed. Two CDC light traps baited with CO_2 were set once a week in the Pearl River Wildlife Management Area (PRWMA), which consists of 6,925 acres, primarily composed of bottomland hardwood forest with wetland areas. The collection area is adjacent to the 1,500 acre waterfowl refuge which is located at the southwestern end of the management area. Weekly collections were made January 2005 through December 2006. Traps were placed mid-afternoon and picked up the following morning. A total of 199,222 mosquitoes were collected during the 2-yr study. No *Cx. quinquefasciatus* were collected throughout the entire study, although other health department surveys have indicated they are abundant just a few miles away. As for other potential WNV vectors, 1,325 (0.6%) *Cx. nigripalpus*, 1,804 (0.9%) *Cx. restuans*, and 6,076 (3.1%) *Cx. salinarius* were collected in the PRWMA over the 2-yr period. These data suggest that *Cx. quinquefasciatus* is not usually found in remote forested environments, but is more associated with human habitation.

P-12 Prevalence of *Ixodes scapularis* tick-borne pathogens in the Lehigh Valley region of Pennsylvania

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Human pathogens transmitted by blacklegged ticks, *Ixodes scapularis*, include *Borrelia burgdorferi* (Lyme disease), *Borrelia miyamotoi* (tick-borne relapsing fever), *Anaplasma phagocytophilum* (human granulocytic

anaplasmosis), and *Babesia microti* (human babesiosis). *Ixodes scapularis* also transmit pathogens that infect animals but not humans, such as *Babesia odocoilei* and specific strains of *A. phagocytophilum*. The extent of the threat to public health from tick-borne pathogens has not been specifically investigated in the Lehigh Valley region of eastern Pennsylvania, population 821,000, where reported cases of Lyme disease are increasing and 3 cases of babesiosis have been identified. Anaplasmosis and relapsing fever are found in neighboring states. Immature ticks acquire *B. burgdorferi* predominantly by feeding on infected white-footed mice (*Peromyscus leucopus*). Eastern chipmunks (*Tamias striatus*) and short-tail shrews (*Blarina brevicauda*) are also competent hosts. Meadow voles (*Microtus pennsylvanicus*) provide blood meals to ticks but are less competent reservoirs. Small mammals serve as sentinels for Lyme disease and are important as primary hosts in the life cycle of blacklegged ticks. This study investigated the infection rates of 4 human pathogens in 100 field-caught rodents and 460 adult and nymphal blacklegged ticks using real-time PCR assays. This work will serve as a baseline for future research of new emerging tick borne pathogens in the Lehigh Valley region.

P-13 Withdrawn

P-14 Analysis of mosquito species abundance over the thirteen years West Nile virus has been present in South Dakota

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Since the 2003 epidemic outbreak of West Nile virus (WNV) in South Dakota, mosquito surveillance has become an integral part of monitoring and predicting WNV occurrence. This year, over 70 CDC CO_2-baited photoreceptor light traps were used across the state to monitor mosquito populations and collect samples for WNV testing. Consistent trapping methods have been used at one trap site in Brookings, SD, since 2001, which gives a unique perspective on the changing dynamics of mosquito populations since that time. This site is located on the outskirts of the city and uses current mosquito collection traps. The trap is activated at dusk and deactivated at dawn the next morning. Samples were collected daily and specimens identified to species. The results showed that *Culex tarsalis* Coquillett has some variation in abundance between years, but remains the most abundant vector mosquito overall. Two other vectors, *Cx. pipiens* L. and *Cx. restuans* Theobald, are collected every year and can vary in abundance from < 1% to nearly 10% of the population. In the past few years, an additional competent vector, *Cx. salinarius* Coquillett, has been collected from the traps, increasing from 0.03% abundance in 2013 to 1% in 2014. Yearly and monthly temporal abundance comparisons for all species collected will be discussed as well as comparisons to other local trapping sites.

P-15 Possible range expansion of *Aedes aegypti* in Palm Beach County, Florida

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Disease transmission is directly tied to the spatial distribution of disease vectors. The range of *Aedes aegypti* in the United States has diminished since the introduction of *Ae. albopictus* in the 1980s; however, *Ae. aegypti* persists in some urban areas, particularly in south Florida. We studied the spatial patterning of *Ae. aegypti* at a fine landscape scale by comparing the distribution of *Ae. aegypti* in Palm Beach County, FL from 2006-2007 to the distribution from 2013, taking into account microclimate and land cover. We found landscape and microclimate factors help explain the distribution of the 2 mosquitoes with evidence for a local range shift of *Ae. aegypti*. This local change in distribution may have implications for shifts on a much broader scale, with concomitant changes in risk of disease transmission.

P-16 Evaluation of two options for molecular detection of West Nile virus in mosquito pools: real time RT-LAMP and real time RT-PCR

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Reverse-transcriptase polymerase chain reaction (RT-PCR) is a well-known method used to detect arboviral RNA in mosquito pools. Reverse-transcriptase loop-mediated isothermal amplification (RT-LAMP) is a method of nucleic acid amplification that can serve as an alternate assay to RT-PCR. Like RT-PCR, RT-LAMP uses a reverse transcription step to synthesize DNA from an RNA template, and then exponentially amplifies DNA to detectable levels in a single-tube reaction. However, the amplification mechanisms are very different affording each assay unique characteristics that should be taken into consideration when choosing the assay that will best serve an agency's needs. We evaluated the sensitivity and specificity of a commercially available West Nile virus (WNV) RT-LAMP assay (Pro-AmpRT™ WNV; Pro-Lab Diagnostics, Inc.) and compared the results to the real-time RT-PCR assay most commonly used by laboratories to detect WNV RNA in mosquito pools. Several types of samples (serial dilutions of WNV, mosquitoes intrathoracically-inoculated with WNV, and field-collected mosquito pools) were tested by both assays. The sensitivity and specificity of both assays were comparable. Practical characteristics of each assay will also be presented including cost, time, reagent and equipment requirements, and other considerations.

P-17 Ecology of La Crosse virus vectors along a forest-to-field ecotone in western North Carolina
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La Crosse encephalitis (LAC) is a pediatric disease that has seen a recent emergence in the number of reported cases in Appalachia. The LAC virus (LACv) is historically transmitted by the sylvan eastern tree-hole mosquito (*Aedes triseriatus*). However, recently invasive species (*Ae. albopictus* and *Ae. japonicus*), which are likely secondary peri-domestic vectors known to co-occur with the native species, complicate the understanding of LACv ecology.

The goal of this study was to evaluate the effect of landscape structure and artificial containers on the distribution and abundance of LACv vectors along a forest-to-field ecotone. We hypothesized a difference in the vector distribution along the ecotone, with the sylvatic species mainly inhabiting the forest and the invasive anthropophilic species inhabiting the more open field or edge habitats. Introducing artificial containers into the field was expected to pull both native and invasive species further into the field and forest, respectively. We ran 2 parallel transects per site (6 sites total), each 200 m in length, 15 oviposition-traps per transect. We also deployed traps for gravid and resting mosquitoes. We incorporated 9 tires in each experimental plot: 2 sites received treatment in the field, 2 sites in the forest, and 2 sites served as control.

Preliminary results suggest habitat preferences with *Ae. albopictus* more abundant in the field habitats, and *Ae. japonicus* as well as *Ae. triseriatus* preferring the forest-edge habitats. The artificial container introduction appeared to increase the abundance of all species, particularly in their "preferred" habitats; however, it did not result in altered oviposition patterns along the ecotone.

P-18 Effects of forced egg retention on *Aedes aegypti* (Diptera: Culicidae)
Monica Montiel, mdmontiel@utpa.edu and Christopher Vitek

Egg retention by gravid female mosquitoes is the by-product of decreased oviposition habitats, either in response to drought conditions or habitat reduction through increased mosquito control efforts (such as container dumping). Forced egg-retention has been reported to have a strong influence on the biology and behavior of *Aedes aegypti* mosquitoes. Research has shown that egg retention may influence adult physiology, oviposition patterns, and offspring development. However, little research has been done on the effect of forced egg retention on the offspring and their subsequent behavior. In addition, forced egg retention may result in different behavior in mosquito populations that are frequently exposed to drought conditions, such as seen in South Texas. We examined the effects of forced egg retention on mosquito offspring. Mosquitoes from an F2 generation were reared in colonies and allowed to oviposit 1 wk after blood feeding (control group) or at increasing intervals of time following blood feeding (treatment groups). The effect of increasing egg retention on oviposition rates, egg survival, hatch rates, development times, and female size of the offspring were recorded. In addition, potential generational effects on future offspring were examined by recording the oviposition rates, hatch rates, and larval survival for the subsequent F3 generation. Understanding the potential response of *Ae. aegypti* to drought and forced egg retention may increase our ability to predict timing of mosquito population increases and increased risk of disease transmission.

P-19 Temporal changes in *Aedes aegypti* and *Aedes albopictus* (Diptera: Culicidae) in South Texas
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Dengue virus and chikungunya virus can be transmitted by 2 species of mosquitoes, *Aedes albopictus* and *Ae. aegypti*, although with different efficiencies for each species and each virus. South Texas represents a potential location for introduction of these viruses. Previous dengue outbreaks in northern Mexico spread into South Texas during peak transmission periods. With the introduction of chikungunya virus to the Western Hemisphere, this virus remains a threat for introduction from Mexico as well. Understanding population dynamics and interactions between these 2 species can increase our ability to estimate risk of transmission for either or both of these viruses. We examined temporal variation of these 2 mosquitoes over the peak activity periods of both of these mosquitoes in 2010 and 2013. During both years, we saw a seasonal decrease in the relative abundance of *Ae. aegypti* compared to *Ae. albopictus*, despite increasing temperatures and decreasing humidity. In 2013, this decline was documented in residential habitats, but not in tire shop sites or cemeteries. This trend may indicate a differential risk of chikungunya virus and dengue virus in South Texas, relative to the time of year and expected relative abundance of either mosquito species.

P-20 Competition between two strains of container mosquitoes in South Texas, USA
Sakshi Puri, sakshi.puri@hchd.org and Christopher Vitek

Dengue virus is transmitted primarily by *Aedes aegypti* mosquitoes, with *Ae. albopictus* being a secondary vector. In 2013, the Texas health service reported 17 cases of dengue in Hidalgo and Cameron County, TX, USA, with at least 2 cases being endemic. Understanding factors that influence vector distribution,

abundance, and growth enable increased accuracy of reporting dengue risk in a region of the country that is historically underserved and potentially at risk. It also allows for more effective control measures to mitigate or eliminate this risk. The majority of research on these species in the USA has been conducted in Florida, and strains that are local to that region. However, South Texas, while also a sub-tropical habitat, is a very different habitat, with increased temperatures and decreased humidity. Patterns of development and emergence due to competition between these 2 vectors that have been identified in Florida may be vastly different in South Texas. We reared F2 generation *Ae. aegypti* and *Ae. albopictus* at varied densities to examine potential interspecific and intraspecific competition. Two temperatures were studied, 92° (representing summer temperatures) and 74° (representing winter temperatures). We recorded the effect of these treatments on larval survival, development time, emergence rate, and adult size. F2 generation females were allowed to oviposit, and the potential effects were also recorded for the F3 generation larvae. The implications of these results of dengue risk and vector control in the region are discussed.

P-21 A three year seasonal survey of adult mosquitoes in a city park
William Dees, wdees@mcneese.edu, Caleb Ardizzone, Jill Hightower, Taylor Wood, Alan Shudes and Terry Sylvester

A seasonal longitudinal survey of adult mosquitoes is underway at a newly renovated, residential park. The total area of the woodland park is 24 acres. The park is separated into 2 distinct sections: one is an open area with playground equipment, picnic tables, open shelters, a small conference center, exhibits, wetland ponds, and concrete walking paths with benches, the other is a preserved forest with nature and hiking trails. The mosquito survey was initiated in summer 2011. We use Centers for Disease Control and Prevention (CDC) light traps baited with CO_2 to collect mosquitoes. Mosquitoes were collected in the open area near the preserved forest in each of the 4 seasons: spring, summer, fall and winter. Meteorological data were also recorded during each trap night. The predominant species collected in a trap night (i.e., >50 mosquitoes) were *Aedes taeniorhynchus*, *Ae. vexans*, *Coquillettidia perturbans*, *Culex nigripalpus*, *Cx. salinarius*, and *Psorophora columbiae*. *Mansonia titillans* and *Uranotaenia* spp. were collected only in the fall. *Culiseta inornata* and *Cq. perturbans* were collected in the winter and spring, respectively. Information obtained in this study can be used to determine potential health risks associated with nuisance and disease vector species. Data from this study also will broaden our understanding of mosquito population dynamics. Studies such as this are excellent for introducing undergraduate students to field research.

P-22 Oviposition preferences of tree hole mosquitoes in an urban mosaic habitat
Gideon Wasserberg, g_wasser@uncg.edu

Natural variation in oviposition site selection has been documented in mosquitoes, and this has been complicated by the introduction of invasive species, resulting in interspecific competition and habitat segregation. In the primarily sylvan *Aedes triseriatus* and the ubiquitous *Ae. albopictus,* oviposition habitat preferences have been established, yet vertical and horizontal components across habitats have largely been overlooked. The characterization of these aspects of vector biology will likely have serious implications on disease management. This study seeks to examine *Ae. albopictus* and *Ae. triseriatus* ovipositional distributions across varying heights and habitats. In accordance with previous literature, we hypothesized that the more urbanized *Ae. albopictus* would oviposit more frequently at low heights in areas characterized by high anthropogenic disturbance while *Ae. triseriatus* would preferentially oviposit in low understory levels of forested areas. Oviposition activity of *Ae. albopictus* and *Ae. triseriatus* was monitored through the deployment of oviposition cups at different heights (0 m, 4.5 m, 9 m) in oak trees across different habitats (forest, field, urban) on the UNC-Greensboro campus. Three trees in each habitat were selected and sampled for 8 consecutive wk in 2013. Linear regression analyses found no significant interactions between species and height or species and habitat type, but oviposition activity for both species increased with canopy cover. An inverse relationship was documented for height and oviposition activity. Overall, *Ae. albopictus* larvae raised from oviposited eggs comprised over 70% of total larvae collected.

Education

P-23 The Mississippi Mosquito and Vector Control Association: Celebrating 25 years of progress in improving mosquito control in Mississippi
Jerome Goddard, JGoddard@entomology.msstate.edu, Nyki Preacely, Sharon Sims, Tina Nations and Wendy Varnado

In 1989, 10 people from throughout Mississippi interested in improving quality and safety of mosquito control created a statewide "Mosquito Control Commission (MCC)," based on a Mississippi law (§41-27-1). These 10 people, representing the Health Department, Mississippi State University, the Mississippi Department of Agriculture, a mosquito products vendor, a county mosquito control commission, and 1 private consultant, met on 3 occasions to develop a broader association of persons interested in mosquito control in Mississippi. Four individuals are primarily responsible for spear-heading a statewide MCC – Dr. Jim

Hamer, an extension entomologist at Mississippi State University, Mr. Harry Fulton, from the Mississippi Department of Agriculture, Dr. Johnny Ouzts, of Delta State University, and Mr. Hoot Holiman, of ADAPCO, a mosquito control products vendor. In March 1990, the first ever Mosquito and Vector Control Workshop was held in Jackson, MS, which consisted of 13 presentations and was attended by over 100 people from around the state. On July 5, 1990, the Mississippi Mosquito and Vector Control Association (MMVCA) was officially registered with the Mississippi Secretary of State. Since then, the MMVCA has flourished with a membership of greater than 150 individuals representing diverse backgrounds in mosquito and vector control, public health, agriculture, and government. For 25 years MMVCA has excelled annually in educating and training mosquito control personnel through vector control workshops, educational material development, and community outreach for Mississippians throughout the state.

GIS/GPS

P-24 Operational use of GIS technology in mosquito surveillance and control within the Salt Lake City District

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Effective surveillance should be the backbone of any professional mosquito and vector abatement district. With growing concerns for economic, environmental, and social issues in relation to the use of pesticides, mosquito abatement districts should strive to continually increase their accountability, record keeping and record sharing. The use of geographic information systems (GIS) has allowed Salt Lake City Mosquito Abatement District to better capture, store, and analyze field collected surveillance data, while also leading to more efficient, effective, and timely control applications. We are primarily utilizing ArcGIS online for most rural operations within the district, and TerraSync for most urban applications. Some components of our operations include site mapping for aerial control measures, inspection data, treatment records, photo collection of sources where appropriate and the ability to quickly sync that data for review and further instruction by management. Our poster will highlight some components of our GIS capabilities and will allow the audience an opportunity to browse through our hardware and software for an interactive experience with our system.

Larval Control

P-25 Multi-site assessment of potential effects of VectoBac™ WG on non-target invertebrates in wetlands distributed along the French Atlantic coast

Laurent Lagadic, laurent.lagadic@rennes.inra.fr, Sebastien Chouin, Marc Roucaute and Jérôme De Maupeou

French regulation requires that, when implemented in protected natural areas (e.g., the European network Natura 2000), mosquito control programs include the assessment of environmental impacts on non-target fauna. Since 2011, 5 sites representing dominant priority endangered habitats along the French Atlantic coast have been monitored for potential effects of VectoBac™ WG on non-target invertebrates. These sites correspond to brackish and fresh waters where mosquito larvae develop. Control and treated areas (several hundred-m² each) were delimited in each site. Five replicate samples were taken in each area, at regular time intervals, up to 5 to 6 sampling dates per year. Water quality parameters were measured at each invertebrate sampling date, so that environmental homogeneity between control and treated areas could be tested, and potential effects of VectoBac™ WG could be discriminated from natural fluctuations of organism abundance. Non-target invertebrate communities were described using diversity and richness indices, and comparisons between treated and control areas were performed using similarity indices and principal response curve. The results show that, over 4 yr, no significant change in invertebrate abundance and diversity could be attributed to VectoBac™ WG applications made according to good mosquito control practices (i.e., at recommended rates). Fluctuations in invertebrate communities were rather associated with changes in environmental conditions, mainly water level. No change was observed for invertebrates which can be used as food sources by predator vertebrates, so that alteration of trophic chains by VectoBac™ WG is very unlikely.

P-26 Gauging efficacy of auto-dissemination stations for *Aedes albopictus* control

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Suppression of *Aedes albopictus* populations is a challenge for mosquito control programs because immatures of the species are found in numerous kinds of peridomestic artificial containers that are difficult to detect, access, and eliminate. *Aedes albopictus* also uses cryptic habitats such as corrugated extension gutters, fence pots etc. as larval habitats, which were invulnerable to conventional control methods. Such behavior makes this species difficult to monitor and manage. This study was designed to assess the feasibility of using auto-dissemination stations (ADS), especially to target the *Ae. albopictus* larval habitats

in urban areas. The pull-and-push idea was to attract wild gravid females to ADS, contaminate them with pyriproxyfen (juvenile hormone analogue), and have them disseminate this insect growth regulator to larval habitats using their skip oviposition behavior. We used oviposition cups to monitor *Ae. albopictus* populations in 3 sites treated with ADS and 3 untreated sites (~3 acre each site) in Mercer County, NJ. We observed significantly lower average daily egg counts in the treatment sites (Wald $\chi 2 = 42.6$, $P < 0.0001$). Our data suggested that using ADS as an element of integrated management program for *Ae. albopictus* is worth further investigations.

P-27 Crowding effects in the yellow fever mosquito (*Aedes aegypti*) and its impact on insecticide sensitivity

Thomas Bilbo, thomas.bilbo@ttu.edu, Dan Dawson and Chris Salice

Aedes aegypti is a globally important mosquito species because it is a principle vector of dengue, yellow fever, and chikungunya. In order to better control *Aedes aegypti* populations it is necessary to thoroughly understand its ecology, life history traits and response to control methods. Larval density is important because it has strong effects on survival and future fitness. Density effects can result from resource competition or crowding, the latter of which has received less research attention despite its ecological importance. Both physical and chemical components cause crowding interactions in larval mosquitoes, which result in increased mortality, prolonged development, and reduced size. The objectives of this research were to determine how different crowding conditions affect insecticide sensitivity. I hypothesized that stress due to crowding would increase insecticide sensitivity. Preliminary results indicate that when larvae are reared at various crowding densities (without resource competition), but later exposed to insecticide at equal densities, they exhibit similar sensitivity. However, when larvae were reared at equal densities, but exposed at various crowding densities, there appears to be a protective effect of crowding, as more densely crowded larvae were significantly less sensitive. These results are somewhat counterintuitive and future efforts will determine underlying mechanisms of observed effects. This research provides important insights into how mosquitoes may respond to control efforts as well as providing empirical recommendations on designing laboratory toxicity tests to better reflect ecological conditions in natural mosquito populations.

P-28 Treating water in cracked soils using an aerial application

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The Coachella Valley Mosquito Control District includes the north end of a large inland saltwater lake known as the Salton Sea. The Salton Sea is fed by agricultural run-off channels and has been receding. The soft sandy and silty clay loams are difficult to walk on, much less to drive equipment on, and technicians have found that they have sunk into the soil as they try to walk. Further, as land that was once inundated becomes exposed, the soil has cracked, and standing water is present in areas 12 inches below the surface. *Culex* larvae have been found living in the water in the cracks, and treating those larvae has been challenging. We evaluated the use of a liquid application of VectoLex WDG and VectoBac WDG using MicroNair AU 7000s mounted to a helicopter. Water sensitive cards showed that droplets were reaching the bottom of the cracks. We also placed 20 5 oz Dixie cups and 10 15-mL vials in the cracks at random locations to determine if enough product was present to control mosquitoes. When returned to lab, water and lab-reared larvae were added to the containers. All larvae were killed within 24 h. We believe that this application method will prove useful to sites where gaining access is difficult.

Operations

P-29 Pre-detection pilot surveillance and outreach program informs post-detection response plan for invasive *Aedes* (*Ae. albopictus, Ae. aegypti*) mosquitoes

Ada Barros, adab@placermosquito.org and Mary Sorensen

Since 2009, invasive *Aedes* mosquitoes (*Ae. albopictus, Ae. aegypti, Ae. notoscriptus*) have established at least 8 separate populations in California. Recognizing that these populations continue to expand into areas previously believed uninhabitable by these *Aedes* species, our agency created a program of surveillance and community outreach to prepare for a possible future invasion of *Aedes*. The program strategy was to 1) identify target sites based on habitat suitability and potential routes of introduction; 2) develop a communication plan and establish communication with stakeholders; and 3) carry out surveillance with ovitraps and larval inspections. While invasive *Aedes* species were not found during our 2014 program, the methods used served the purpose of preparing our agency, training our staff, and educating the community about the threat of invasive *Aedes* mosquitoes.

P-30 Out-of sight, out-of-mind: mosquito production in subsurface stormwater infrastructure *The Infectious Disease Dragon Under Our Feet*

Amber Semrow, asemrow@ocvcd.org, Robert Cummings, Kiet Nguyen and Tim Morgan

Underground storm drain systems and other subsurface stormwater infrastructure continue to be prolific mosquito sources (primarily *Culex quinquefasciatus*) during warm months due to persistent dry weather flows in urban/suburban settings. Despite published studies on this issue, public health officials are still calling for greater recognition and collaboration from the stormwater community to address this serious public health threat. Mosquitoes often seek underground sources of standing water in subsurface infrastructure to lay their eggs and use as resting sites. Mosquitoes in this unseen system can be a huge factor in the spread of mosquito-borne diseases, such as West Nile virus (WNV), as was shown during the 2014 epidemic in Orange County, CA, when early season mosquito infection rates in underground sites were much higher than above ground sites. Dry weather flow into these systems from landscape over-watering is arguably the greatest influence in exacerbating subsurface mosquito production. Partnerships which raise awareness about the relationship of water quality and conservation are commonplace; however, the public health component is largely missing from the dialogue. Water misuse and poor water quality increases the risk to the public of mosquito-borne disease. It is incumbent on the stormwater and mosquito control communities to work synergistically to reduce conditions that favor mosquito oviposition in stormwater infrastructure. This reduces the need for pesticide applications, expensive retrofits, and abatement costs. The integration of mosquito reduction measures into relevant facets of stormwater management is a "win-win" for improving water quality and public health protection.

Systematics

P-31 Identification keys for biosurveillance of *Anopheles* and *Culex* mosquitoes in Africa

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The Walter Reed Biosystematics Unit (WRBU), Entomology Branch, WRAIR is developing interactive, computerized identification keys for adults and larvae of mosquitoes, as major tools for the biosurveillance of disease vectors and related species. Using LUCID software, identification keys for the Afrotropical Region have been created, primarily for genera *Anopheles* and *Culex*, including vectors and related species. For genus *Anopheles* of the whole African region, more than 100 species were treated in separate adult and larval morphological keys. For genus *Culex* of West, Central and East Africa, more than 30 species in subgenera *Culex* and *Oculeomyia* were treated for adult identification keys. Diagnostic characters of the head, thorax (including legs and wings), and abdomen were selected and scored in matrix format for use in the LUCID keys. Automontage images of contrasting states for each character were prepared, labeled and included in the keys. Species pages of known or potential disease vectors in *Anopheles* and *Culex* were also completed, with brief notes on the taxonomy (including synonyms or other invalid names), bionomics, medical importance, geographical distribution, selected images of habitus and other body parts, and relevant references, with links to WRBU/AFPMB LRS mosquito databases (pdfs, etc.). Mosquito catalogs were noted, including their continuous updates for species of *Anopheles*, *Culex* and other mosquito genera from Africa and other regions of the world.

P-32 Withdrawn

P-33 Developing MALDI-TOF MS method for routine identification of mosquito species occurring in Canada

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Mosquito identification is typically based on morphological features using taxonomic identification keys and the process is labor intensive. In addition, not all specimens can be accurately identified because critical morphological structures can be lost from specimens during collecting and sorting. The use of protein profiling by matrix-assisted laser desorption ionization-time-of-flight mass spectrometry (MALDI-TOF MS) has been increasingly used for the routine identification of bacteria, and recently for arthropods. The method is accurate, fast and inexpensive after preparing the MALDI spectral database. We created a database of reference spectra at the National Microbiology Laboratory, Winnipeg, Manitoba after testing protein extracts from mosquito legs. So far, the database includes more than 30 species from *Aedes*, *Ochlerotatus*, *Culex*, *Anopheles*, *Culiseta*, *Coquillettidia*, *Uranotaenia*, and *Psorophora* genera. An overview of this novel process will be provided as well as a discussion of the specificity of the approach.

Other

P-34 Depletion of amino acids, acylcarnitines and organic acids leads to permethrin resistance in *Aedes aegypti* in Mexico

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Aedes aegypti, the principal vector of dengue, occurs mainly in tropical and subtropical areas. The application of insecticides is a major tool in reducing dengue transmission. Pyrethroids have been commonly used due to their high toxicity to target insects; however, after decades of application the increasing reports of resistance compromise the effectiveness of control programs. Insecticide resistance is associated to a metabolic and energetic cost that can influence mosquito biology in the field. Fitness cost can be considered a consequence of trade-off between the allocations of energy insecticide resistance mechanisms, affecting both developmental and reproductive parameters. The energetic cost in *Ae. aegypti* never has been studied with insecticide resistance. We attempt to determine if there is a fitness cost related with metabolites reduction in insecticide-resistant population. A field population of *Ae. aegypti* was exposed to a diagnostic dose (DD) of permethrin (15 µg/ml) at diagnostic time (30 min) using bottle bioassay. Survivors were collected and compared with non-exposed individuals and the susceptible New Orleans strain. By tandem mass spectrometry, we obtained the reference ranges concentrations of amino acid (AA), acylcarnitines (AC), and organic acids (OA). The means and standard deviations were determined and statistically significant differences were observed between groups. The levels of short, medium, and long chain AC, AA, and OA were significantly different. Ranges of AC, AA, and OA in *Ae. aegypti* from 2 strains (susceptible and resistant without exposure to insecticides and survivors to a DD of permethrin) were also determined and significant differences were found.

P-35 Analysis of population structure and insecticide resistance in mosquitoes of the genera *Culex*, *Aedes* and *Anopheles* from different environments in N. Greece

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The re-introduction of vector-borne diseases in countries such as Greece where they had been eradicated for many years is a major public health problem. Use of insecticides is the primary means for control. Knowledge of population ecology and insecticide resistance is a pre-requirement for the application of appropriate control strategies. We are studying the population dynamics, presence and mechanisms of insecticide resistance in mosquito populations of the genus *Anopheles* (e.g., *An. sacharovi*, *An. hyrcanus* and *An. maculipennis* – possible malaria vectors), *Culex pipiens* (West Nile virus vector) and *Aedes caspius* (major nuisance mosquito). By using modern molecular diagnostic tools for species ID markers and target site resistance mutations, as well as CDC bioassays and biochemical tests for metabolic resistance, we examine and compare the species composition and the insecticide resistance status of these populations in light of the support and optimization of mosquito control and resistance management strategies.

P-36 Evaluating the impact of stormwater mitigation on mosquito populations in New Orleans, Louisiana

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New Orleans is currently experiencing a rapid increase in the development of green infrastructure projects to mitigate stormwater and to address its unique drainage and flooding issues. During rainfall events, pumps are required to remove water from low-lying neighborhoods. Local rain gardens aim to act as water divergence systems and temporary storage for municipal stormwater. In order to determine if these 4 rain gardens also act as mosquito oviposition sites , weekly inspections and surveillance utilizing gravid traps baited with a hay and fish oil emulsion were conducted. Samples were tested for arboviruses such as West Nile and St. Louis encephalitis viruses via RT-PCR. Surveillance efforts also incorporated CDC light traps and BG Sentinel® traps to target additional mosquito species. Preliminary results show focal differences in population magnitude and species composition between rain gardens. Better knowledge of the potential role of rain gardens and mosquito production will help inform city officials, stakeholders, and the general public on future stormwater mitigation strategies.

Latin American

P-37 The measure of intensity of resistance in *Aedes aegypti* to pyrethroids using the enhanced surveillance protocol for the CDC bottle bioassay

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Insecticides play an important role in vector control programs. However, local information about resistance intensity from different regions is needed to make a decision about which insecticide or insecticides may be applied. In this context, the aim of the present study was to determine the resistance frequency through the bottle bioassay protocol conducted upon *Aedes aegypti* female mosquitoes from 3 different regions of Mexico. Up to 25 mosquitoes per assay replicate (up to 4 replicates, and a control group) were exposed to the diagnostic insecticide dose (DD) and 2, 5 and 10 times the DD at a certain diagnostic time of 3 pyrethroids (permethrin, bifenthrin and *d*-phenothrin). The comparison of the intensity of resistance among pyrethroids will directly measure the potential for differential toxicity to allow more sustainable use of different subclasses of pyrethroids. Also, the enhanced surveillance protocol used in populations within different sites of a region could be a reference to allow decision-making based on the significance of resistance at varying frequencies and intensities of individual insecticides, in combinations and/or rotations.

P-38 Effects of synergists on toxicity of permethrin in *Aedes aegypti* (Diptera: Culicidae)

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The resistance to and the effects of synergists on the toxicity of permethrin in *Aedes aegypti* were studied. Piperonyl butoxide, (PBO), S,S,S,-tributylphosphorotrithioate (DEF), triphenyl phosphate (TPP), and diethyl maleate (DEM) were chosen to be applied in a susceptible as well as in permethrin resistant populations. Significant synergistic effects on permethrin toxicity were found by using DEF. DEM and PBO show little synergistic effect and the resistance levels remained high even with the presence of TPP. The resistance levels to permethrin could be inhibited strongly by applying DEF. From the results, esterase degradation is believed to play a critical role in resistance to permethrin in *Ae. aegypti.* To a lesser extent, hydrolytic reactions also were partially involved in the resistance to permethrin by using the synergists PBO and DEM.

P-39 Autodissemination of entomopathogenic fungi as alternative control of house fly (*Musca domestica* L).

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Musca domestica L. is perhaps the best icon of anthropophilia capable of transmitting multiple pathogens to humans and animals. Chemicals are often used to reduce fly populations . However *M. domestica* has been resistant to multiple insecticides since the 1950s. This has given impetus to the exploration of biocontrol approaches such as entomopathogenic fungi. Autodissemination of fungi has been well documented in *Glossina morsitans* and *Aedes aegypti* but not in *M. domestica*. Because *M. domestica* females are monogamous and the males polygamous, autodissemination can be considered. *Beauveria bassiana* and *Metarhizium anisopliae* were autodisseminated in *M. domestica* using 1, 5 and 10 virgin, 3-day old males firstly exposed to 6 X 108 conidia mL-1 per fungus and confined with 30 females of the same age. Fungal treatments with 1, 5, and 10 males caused low mortality with LT_{50} of 7, 5, and 3 days respectively with a sporulation rate of 94% and an oviposition reduction of 66%. The regression of the mean of eggs, first instar larvae and days of gonotrophic cycle (GC) on the LT_{50} values of 9 treatments showed that the 3 variables declined in about 3-4 units per unit of LT_{50}. Likewise, when the GC time was regressed on the mean oviposition, a linear equation demonstrated that for each decrease of 15 eggs there was a shortening of 1 day in the GC, which therefore was shortened from 7 days in healthy flies to 2 in females of the fungal treatment with 10 males. This autodissemination could be used to improve the *M. domestica* integrated control in the field and human environments.

P-40 Susceptibility of housefly (*Musca domestica* L.) to commercial insecticides in dairies of Michoacan, Mexico

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Different pest insect and mite species affect dairy industry such as the housefly within which highlights housefly (*Musca domestica* L.) The activity of this pest can decrease dairy production and the feed conversion efficiency in addition to carrying a large number of pathogens, which cause damage to both humans and cattle. One strategy to control this pest is the use of chemical insecticides; nevertheless, the continuous use of this can lead to resistance. There is a chance that in the occidental dairy region of

Michoacan there exists a house fly resistance problem, which contaminates the environment and the final product (milk) causing human intoxications. The study objectives were to determine the susceptibility of the housefly population to commercial insecticides used in the region. For this, bioassays were developed using adult insects in laboratory conditions which were exposed to treatments by 3 different methods (topical application, ingestion and residual treatments). LD_{50} and LT_{50} were determined to define the concentration of doses to field application. According to the results, cypermethrin at doses of 2.5, 5, 10 and 20 ml/L has a mortality of 90, 80, 96.7 and 96.7% respectively. By demonstrating a high susceptibility, however DDVP, chlorpyrifos ethyl, thiametoxam, deltamethrin, permethrin and fipronil at least in 1 application method is observed that there is less than 66% mortality, which shows that there is possible resistance to these insecticides in the region.

P-41 Mosquito diversity (Diptera: Culicidae) in Los Tuxtlas region, southern Veracruz, Mexico

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A study of culicid mosquito diversity in the region of Los Tuxtlas, Veracruz, was based in the collection of adults in May, September, and February, 2010-2011. Three study sites , 2 with preserved environments (Cerro del Borrego, Municipality of San Andrés Tuxtla and Cerro Pipiapan, municipality of Catemaco) and 1 urbanized (Montepío, municipality of San Andrés Tuxtla), were used to compare the differences in species richness and relative abundance, analyzed by using biodiversity measurements as proposed by Magurran (2004) and Moreno (2001). A total of 1,124 individuals belonging to 9 genera and 32 species were obtained, representing 23.02% of the species historically recorded for the state of Veracruz. The sampling site with the most diversity was the urban environment of Montepío (D = 4.16; H ' = 1.7), since ensemble showed low dominance and high equitability as compared with the other 2 sampling sites. Seasonal weather variation influenced mosquito diversity; rainy season (September sample) was the time of the year in which higher relative abundance and species richness probably as response of the presence of any kind of natural and artificial water collections useful for immature development. The species inventory generated in this study provides updated and useful information of the mosquito species, some of which are well known vectors of viral and/or parasitic zoonotic diseases; 10 species found in the study area should be considered important for Los Tuxtlas public health surveillance programs: *Aedes scapularis, Ae. taeniorhynchus, Ae. triseriatus, Ae. angustivittatus, Anopheles albimanus, An. pseudopunctipennis, Culex nigripalpus, Cx. quinquefasciatus, Haemagogus mesodentatus* and *Mansonia titillans* with orthologous genes from others mosquitoes.

P-42 Characterization of *Aedes aegypti* oviposition sites in Medellin, Colombia

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Aedes aegypti is the vector of dengue fever in Medellin, one of the most densely populated cities in Colombia, where the disease is endemoepidemic. Despite different efforts to generate a vaccine, currently, the most appropriate anti-dengue strategy is reducing the vector population, and the elimination of larval habitat is the action that generated the best long-term results. However, for this type of anti-vector action to be appropriate, it is necessary to know the availability, preference and productivity of different types of larval sites. This study was conducted to characterize *Ae. aegypti* oviposition sites in Medellin and provide relevant information for decision-making. To characterize larval sites, information was analyzed from traditional mapping of entomological indices (EI) carried out in the last 7 yr. The EI in Medellin are done quarterly, and on each of these occasions, all neighborhoods of the city (252) are visited. The information assessed corresponded to more than 170,000 households, and information from the deposit infestation index was used to determine the most abundant type of site and that which is most preferred by the vector. The analysis was performed on a spatiotemporal scale, and the neighborhood and the quarter in which the EI was performed were used as units of study. Additionally, the abundance and type of oviposition sites were analyzed according to the socioeconomic status of each neighborhood. The results allowed the determination of the spatial and temporal fluctuations in the amount and type of vector production sites. In addition, some sites showed higher preference; however, this was not significant between different socioeconomic strata.

P-43 Frequency of mutations in the acetylcholinesterase, carboxicolinesterase 1D and b-esterase 6 genes in *Aedes aegypti* from Merida, Yucatan, Mexico

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Metabolic resistance to pesticides has been poorly elucidated in the dengue mosquito vector *Aedes aegypti*. Three enzyme families have been involved in the metabolism of insecticides, cytochrome P450 monooxygenases (P450), glutathione transferase (GST), and carboxyl/cholinesterases (CCE). Organophosphate or carbamate resistance in *Ae. aegypti* have not yet been associated with potential detoxification genes. The goal of this study was to generate a chlorpyrifos resistant *Ae. aegypti* strain and determine the allele frequency for mutations at potential candidate genes, including acetylcholinesterase

(ace-1), esterase-6 (est-6) and carboxyl/esterase (CCEae1D). We selected a mosquito strain collected in Hunucma, Yucatan, in Mexico. Three generations of selection were performed with the bottle bioassay using lethal concentrations that kill 50% of the mosquitoes (LC_{50}). A sample of alive and dead mosquitoes was separated after chlropyrifos exposure during each generation. DNA was isolated from ~50 individual mosquitoes per generation and we performed melting-curve PCR to obtain the genotypes for each loci. Allele frequencies were calculated for each gene and compared between alive and dead groups. Chlorpyrifos LC_{50} increased after 2 generations of selection (1.1 fold). Our results suggest that allele frequencies at ace-1 and CCE-1D are not associated with chlorpyrifos resistance in Hunucma, however, allele frequencies in est-6 are significantly higher in chlorpyrifos resistant mosquitoes. Further selection of additional mosquito strains and characterization of these candidate genes will be required to develop a molecular diagnostic test for chlorpyrifos resistance in *Ae. aegypti*.

P-44 Molecular characterization of OR1, OR2, OR7 and OR47, odorant receptors genes from *Aedes aegypti*

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The house mosquito, *Aedes aegypti* perceives the world through small, signal-carrying molecules (semiochemicals). Understanding this mechanism may lead us to discover novel repellents for reducing bites and disease transmission as well as "green chemicals" for monitoring and controlling mosquito populations. The molecular characterization of the *OR* (odorant receptors) genes at the moment is not currently understood. The goal of this study was to characterize the sequence of different transcripts of *OR1, OR2, OR7* and *OR47* genes from *Ae. aegypti*. Larvae samples were obtained from water storage containers around Guadalupe, Nuevo León, and Mexico. Total RNA extraction was accomplished by using a Trizol® reagent while following the manufacturer's instructions. cDNA was synthesized by reverse transcription reaction with high capacity reverse transcription kit using oligo dT. Primers were designed using the online tool Primer3. The amplified products were run in agarose gels and showed bands of expected sizes (1540, 1632, 2454 and 2469 bp), these amplified products were cloned in the plasmidic vector XL-TOPO and then sequenced. The homology of these genes was deduced by comparing them.

P-45 Characterization of four 5α- esterase from *Culex quinquefasciatus* Say (Diptera: Culicidae)

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Esterases have been an important part of insect biochemical research over the last 50 yr. As an important component of insect xenobiotic defense system, they have been a focus for studies of insecticide metabolism and resistance. To date no insect esterase gene has been cloned, but several have been isolated, particularly from *Drosophila.* The goal of this study was to characterize the sequence of different transcripts of 5α- esterase gene from *Culex quinquefasciatus*. Larvae samples were obtained from water storage containers around the metropolitan area of Monterrey, Nuevo Leon, Mexico during 2013. Total RNA extractions were done with Trizol® reagent following the manufacturer's instructions. cDNA was synthesized by reverse transcription reaction with high capacity reverse transcription kit using oligo dT. Primers were designed using the online tool Primer3. The amplified products were run in agarose gels and show 4 distinct bands of 2138, 2079, 2074 and 2024 bp, this mix of bands were cloned in the XL-TOPO plasmid by shotgun and then sequenced. Four species of different transcript from the *Cx. quinquefasciatus* 5α esterase were found. The characterization of this gene could help in future experiments related to the vector control.

P-46 Characterization and quantification esterase from *Aedes aegypti*

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Aedes aegypti, also called yellow fever mosquito, is a mosquito that can spread the chikungunya, dengue fever, yellow fever viruses, and other diseases. Permeability barriers have important roles in many tissues in both vertebrates and invertebrates. In insects, the high potassium concentration of the hemolymph can block action potentials in neurons, and, thus, cause paralysis, if the blood–brain barrier is disrupted. Gliotactin is a serine esterase-like, transmembrane protein expressed in epithelia and glia, and is part of the septate junction complex, which forms the permeability barrier in epithelia and glia. However, in epithelia cells, it is uniquely localized to the corners at the convergence of septate junctions from 3 neighboring cells. This region is known as the tricellular junction (TCJ) and gliotactin is a molecular marker for this junction in *Drosophila*. The TCJ is a specialized region of the permeability barrier and is formed by 3 septate junctions.

In this work, the gliotactin was amplified and the expression levels measured based on different diets. For amplification of the genes specific primers from *Aedes aegypti* were performed based on sequences

obtained from GenBank using primer3 plus program. The amplified products were obtained from design specific primers for the specific detection and quantification. Different levels of gene expression based gliotactin subjected diets (flake fish, liver and yeast powder) were obtained. The results of molecular assays can be used to remove the gliotactin protein results in the disorganization of septate junctions. In glia the septate junctions are also leaky and fail to form the blood-brain barrier resulting in paralysis and death.

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Goddard, Jerome	Mississippi State University, Department of Biochemistry, Molecular Biology, Entomology, and Plant Pathology, Mississippi State, MS, United States	P-23
González-Olvera, Gabriela	Universidad Autonoma de Nuevo Leon., Monterrey, Nuevo León., Mexico	P-45
Graham-Derham, Scott	Manitoba Health, Healthy Living & Seniors, Winnipeg, MB, Canada	129
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Gutierrez-Rodiguez, Selene Marlen	Universidad Autonoma de Nuevo Leon, Facultad de Ciencias Biologicas, San Nicolas de los Garza, Nuevo Leon, Mexico	P-38
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Hardman, Jason	Salt Lake City Mosquito Abatement District, Salt Lake City, UT, United States	40
Harrison, Cynthia	New Orleans Mosquito & Termite Control Board, New Orleans, LA, United States	84
Hartle, Jacob	Clarke, St. Charles, IL, United States	104
Harwood, James	Navy Entomology Center of Excellence, Jacksonville, FL, United States	39
Healy, Kristen	Louisiana State University, Baton Rouge, LA, United States	61, 165
Heft, David	Turlock Mosquito Abatement District, Turlock, CA, United States	17
Henke, Jennifer	Coachella Valley Mosquito and Vector Control District, Indio, CA, United States	190, 221, P-28
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Hogsette, Jerome	USDA-ARS-CMAVE, Gainesville, FL, United States	155
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Howe, Richard	AirWolf Malaria Vector Control Research, Punta Gorda, FL, United States	180
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Markowski, Daniel	Vector Disease Control International, Greenville, MS, United States	21, 41
Matton, Priscilla	Bristol County Mosquito Control Project, Taunton, MA, United States	168
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Moreno-Garcia, Miguel	Department of Microbiology, Immunology and Pathology, Colorado State University, Fort Collins, CO, United States	116

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Rinkevich, Frank	Louisiana State University, Department of Entomology, Baton Rouge, LA, United States	182
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Williams, Gregory	Hudson Regional Health Commission, Secaucus, NJ, United States	109
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Zimler, Rebecca	USDA/ARS, Gainesville, FL, United States	49

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.	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)	2013	Judy Hansen (NJ)
1964	George H. Bradley (USPHS/USDA)	1983	Anthony W. A. Brown (WHO)	2013	Henry Rupp (NJ)

HAROLD FARNSWORTH GRAY MEMORIAL CITATION
MERITORIOUS SERVICE TO MOSQUITO CONTROL AWARD

This now discontinued award was 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 now 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 Medal of Honor is the highest award regularly given by AMCA. The only specific limitation for the Medal of Honor is AMCA membership, and 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

1972	Maurice W. Provost (FL)	1983	Harry D. Pratt (GA)	2001	Gary G. Clark (USPHS)
	William R. Horsfall (IL)		John A. Mulrennan, Sr. (FL)	2002	Lucas G. Terracina (LA)
1973	Don M. Rees (UT)	1984	George T. Carmichael (LA)	2003	Robert J. Novak (IL)
	Thomas D. Mulhern (CA)	1985	Norman G. Gratz (WHO)	2004	James D. Long (TX)
1974	Anthony W. A. Brown (WHO)	1986	James R. Caton (CA)	2005	James W. Robinson (FL)
	Donald L. Collins (NY)	1987	Jay E. Graham (UT)	2006	John L. Clark Jr. (IL)
1975	Daniel M. Jobbins (NJ)	1988	Lewis T. Nielsen (UT)	2007	E. John Beidler (FL)
	Arthur W. Lindquist (USDA)	1989	Andrew J. Spielman (MA)	2008	David A. Dame (FL)
1976	Austin W. Morrill, Jr. (CA)	1990	Glen C. Collett (UT)	2009	Dan Ariaz (NV)
	Carroll N. Smith (USDA)	1991	Harold C. Chapman (LA)		Gary Breeden (VA)
1978	James B. Kitzmiller (FL)	1992	D. Bruce Francy (CO)	2010	Mir S. Mulla (CA)
	William D. Murray (CA)	1993	Gilbert L. Challet (CA)	2011	Dave Brown (CA)
1979	Richard F. Peters (CA)	1994	Ronald A. Ward (MD)	2012	Sammie L. Dickson (UT)
1980	William E. Bickley (MD)	1995	T. Wayne Miller (FL)	2013	Wayne Crans (NJ)
	John N. Belkin (CA)	1996	Marshall Laird (New	2014	Chester G. Moore (CO)
1981	Stanley J. Carpenter (CA)	1997	Robert K. Washino (CA)		
	Roland E. Dorer (VA)	1998	John D. Edman (MA)		
1982	Kenneth L. Knight (NC)	1999	Bruce F. Eldridge (CA)		
	William C. Reeves (CA)	2000	Judy A. Hansen (NJ)		

MERITORIOUS SERVICE AWARD

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 two awards per year.

1972	Charles F. Scheel (IL)	1980	James D. Gorman (FL)	1996	Donald J. Sutherland (NJ)
	Donald L. Collins (NY)		Donald E. Weidhaas (FL)		Ronald A. Ward (MD)
	Theodore G. Raley (CA)		E. John Beidler (FL)	1997	Roger S. Nasci (CO)
1973	Francis P. Creadon (CA)		Eugene J. Gerberg (MD)		Thomas J. Zavortink (CA)
	Vernon Conant (NJ)	1981	A. Ralph Barr (CA)	1998	James D. Long (TX)
	Austin W. Morrill, Jr. (CA)		Gilbert L. Challet (CA)	1999	Hilton B. Munns (CA)
1974	Leslie D. Beadle (USPHS)		Edgar A. Smith (VA)	2000	Leroy J. Bohn (VA)
	John H. Brawley (CA)	1982	Hugo A. Jamnback (NY)		Dreda McCreary (VA)
	John W. Kilpatrick (GA)		Donald R. Johnson (GA)	2001	Charles T. Palmisano (LA)
	T. Oscar Fultz (GA)		Harold D. Newsome (MI)	2002	Thomas G. Floore (FL)
	Howard R. Greenfield (CA)		James V. Smith (GA)		Sherry McLaughlin (TX)
	Paul J. Hunt (FL)	1983	Richard F. Darsie (CO)	2003	Wayne L. Kramer (NE)
	William C. McDuffie (USDA)		Ronald A. Ward (DC)		John L. Clarke, Jr. (IL)
	Donald R. Johnson (GA)	1984	Samuel G. Breeland (FL)	2004	Yadira N. Rangel (Venezuela)
	Helen Sollers-Riedel (DC)		Donald J. Sutherland		James W. Robinson (FL)
1975	Lewis E. Fronk (UT)	1985	John C. Kuschke (NJ)	2005	Major S. Dhillon (CA)
	Joseph G. McWilliams (USN)		James R. Caton (CA)		William H. Meredith (DE)
	Lewis J. Ogden (USPHS)	1986	C. Lamar Meek (LA)	2006	William J. Sames (WA)
	Rajindar M. Pal (WHO)	1987	John C. Combs (CA)	2007	Henry R. Rupp (NJ)
	Kenneth D. Quarterman	1988	Chester G. Moore (CO)	2008	Allan Inman (CA)
	Herbert F. Schoof (USPHS)		Margaret Parsons (OH)		Manuel Lluberas (FL)
1976	Robert A. Armstrong (MA)	1989	John S. Billodeaux (LA)	2009	Joe Conlon (FL)
	Osmond P. Breland (TX)		Edgar S. Bordes, Jr. (LA)	2010	Norbert Becker (Germany)
	George B. Craig, Jr. (IN)	1990	Richard D. Morton (WA)	2011	Harry Savage (CO)
	Claude M. Gjullin (USDA)		Lucas G. Terracina (LA)		L.A. Williams (SC)
	T. Wayne Miller (FL)	1991	David A. Dame (FL)	2012	Lal S. Mian (CA)
1976	Donald J. Pletsch (Mexico)	1992	Jerry Mix (TX)		Edsel M. Fussell (FL)
	Glenn M. Stokes (LA)	1993	William E. Hazeltine (CA)	2013	Kenneth J. Linthicum (FL)
	Luis M. Vargas (Mexico)	1994	Sally A. Wagner (MI)	2014	Diann Crane (MN)
1978	Richard C. Axtell (NC)	1995	Frederick W. Wagner		Daniel Kline (FL)
1979	Marco. E. C. Giglioli (BWI)				

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)	1994	James W. Robinson (FL)	2005	Mark Newberg (IL)
	Leslie E. Fronk (UT)		Dan L. Ariaz (NV)		Susan Maggy (CA)
	Jesse B. Leslie (NJ)	1995	Sally Kuzenski (LA)	2006	Teung Chin
1981	Linda G. Raiche (CA)	1996	Carl R. Tanner (IL)	2007	Karl Malamud-Roam (CA)
	Margaret S. Slater (NY)		Sammie L. Dickson (UT)	2008	Wiliam H. Meredeth (DE)
1982	K. G. Nolan (NY)	1997	Charles T. Palmisano (LA)	2009	Rep. Dennis Cardoza (CA)
	Charles F. Scheel (IL)		George J. Wichterman (FL)	2010	Gordon Patterson (FL)
1983	Coyle E. Knowles (NY)	1998	Douglas B. Carlson (FL)		Gary Clark (FL)
1984	Ray Treichler (DC)	1999	Charles Beesley (CA)		Yasmin Rubio-Palis
1985	Lawrence T. Cowper (USAID)		Donald R. Johnson (GA)	2011	Angela Beehler (WA)
	Janice B. Wells (NY)	2000	Peter B. Ghormley (CA)		Roxanne Connelly (FL)
1986	T. Oscar Fultz (GA)		David A. Brown (CA)	2012	Truc Dever (CA)
1987	Sharon A. Colvin (IL)	2001	Donald Menard (LA)	2013	Robert Peterson (MT)
1988	Daniel D. Sprenger (TX)		Joel Margalit (Israel)	2014	Salvador Rico (TX)
1989	Fred C. Roberts (CA)	2002	Dennis Moore (FL)		
1990	Leonard E. Munsterman (IN)		Henry R. Rupp (NJ)		
1991	James D. Long (TX)	2003	James R. McNelly (NJ)		
1992	Charlie D. Morris (FL)		Robert Bonnett (MN)		
1993	Robert J. Novak (IL)	2004	James R. Brown (FL)		

JOHN N. BELKIN AWARD

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)
1982	Lloyd E. Rozeboom (IL)
1983	Kenneth L. Knight (NC)
1984	Thomas J. Zavortink (CA)
1985	Stanley J. Carpenter (CA)
1986	Elizabeth P. Marks & John Reid (Australia)
1987	James B. Kitzmiller (FL)
1988	Allan R Stone (MD)
1989	Pedro Galindo (Panama)
1990	Peter F. Mattingly (UK)
1991	Jose P. Duret (Argentina)
1992	Bruce A. Harrison (NC)
1993	Edward L. Peyton (DC)
1994	Theodore H. G. Aitken (CT)
1995	Oswaldo P. Forattini (Brazil)
1996	A. Ralph Barr (CA) Michael W. Service (UK)
1997	Christine J. Dahl (Sweden)
1998	Ralph E. Harbach (UK)
1999	Yiau-Min Huang (DC)
2000	Lewis T. Nielsen (UT)
2001	John F. Reinert (FL)
2002	Richard F. Darsie (FL)
2003	Richard C. Wilkerson (MD)
2004	Kazua Tanaka (Japan)
2005	Ronald A. Ward (MD)
2006	William K. Reisen (CA)
2008	Maria-Anice Sallum (Brazil)
2010	Daniel Strickman (MD)
2011	Rampa Rattanarithikul, Ph.D. (Thailand)
2012	Maureen Coetzee, Ph. D. (South Africa)
2013	John F. Anderson (CT)
2014	Graham White (FL)

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 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 & La Crosse 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 Harold F. Gray	Robert K. Washino	
1988	John A. Mulrennan,	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.
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 Management

MEMORIAL LECTURE HONOREE & MEMORIAL LECTURER AWARD (continued)

	HONOREE	LECTURER	TOPIC
2003	Kenneth L. Knight	Ralph E. Harbach	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
2008	Andrew Spielman	John D. Edman	
2009	Lamar Meek	Roxanne Connelly	
2010	Harold C. Chapman	Tokuo Fukuda	
2011	H.G. Dyar	Terry Klein	
2012	James D. Long	John Welch	
2013	Thomas Mulhern	Randy Gaugler	
2014	Founding Mothers of Mosquito Control	Gordon Patterson	

INDUSTRY AWARD

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

1997	Charles T. Galley (FL)	2006	Willie N. Cox (IL)
1998	William German (FL)	2007	Bob Bonnett (MN)
1999	Gary A. Mount (FL)	2009	Clarke Hudson (IL)
	Daniel F. Boyd (GA)		Bill Strange (ID)
	David W. Waldron (GA)	2010	Peter Connelly (FL)
	J. David Waldron (GA)	2011	David Sullivan (MT)
2002	Robert E. Richard (TX)	2012	Stephanie Whitman (WY)
2003	Allen W. Wooldridge	2013	Larry Erickson (IL)
2004	John L. Clarke, Jr. (IL)	2014	Gerry Hutney (FL)
2005	Ernest Danka (IL)		

GRASSROOTS AWARD

This award is given to recognize excellent performance and dedication by mosquito control field staff.

2005	Omar S. Akbari	Reno Washoe Country, Nevada
	Christopher Trapp	Multnomah County Vector Control,
2006	John Phelps	Mercer County, New Jersey
2008	Chris Frame	Cape May County, New Jersey
2009	Jason Craig Hardman	Salt Lake City MAD, Utah
2010	Jessica Fales	Midland County MC, Michigan
	Gary Hillsdale	Metropolitan MCD, Minnesota
	Elizabeth Vice	Buttle County MVCD, California
2011	David Bruget	Kings MAD, California
	Russell Eck	Washoe County Health District, Nevada
	Phillip Henry	Butte County MVCD, California
	Levi Zahn	Williston VCD, North Dakota
2012	Mike Smith	Anastasia MCD, Florida
2013	Arturo Gutierrez	Coachella Valley MVCD, California
2013	Michael Martinez	Coachella Valley MVCD, California
2013	David Lopez	Greater Los Angeles County VCD, California
2013	Martin Serrano	Greater Los Angeles County VCD, California
2014	Dell Boyd	Butte County MVCD, California
	John McCready	Jackson County VCD, Oregon
	Gaby Perezchica-Harvey	Coachella Valley MVCD, California
	Geneva Ginn	Coachella Valley MVCD, California

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

1989	Scott Willis	McNeese State U.
1990	Andrea Brown	Peru State Coll.
1991	John Paul Mutebi	Notre Dame U.
1992	Rosmarie Kelly	U. Massachusetts
1993	Merry L. Holliday-	U. California, Davis
1994	John E. Gimnig	U. California, Davis
	Alice Shaeffer*	U. Mainz, Germany
1995	Glen Scoles	Notre Dame U.
	Jittawadee Rochaeroen*	U. California, Riverside
1996	Esther Chow Schaeffer	U. Maryland
1997	Lynn Cooper	U. Maryland
1998	C. Roxanne Rutledge	Louisiana State U.
	Emmalee Kennedy*	U. Illinois
	Timothy Schaub*	U. Illinois
1999	Laura Harrington	U. Massachusetts
	Adam S. Jones*	U. Massachusetts
	Hillary Reno*	U. Illinois
2000	Jason L. Rasgon	U. California, Davis
	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.
2003	Sarah Yaremych	U. Illinois
	Laura Goddard*	U. California
	Jason L. Rasgon*	U. California, Davis
2004	Gregory M. Williams	U. Delaware
	Stephen Aspen*	Colorado State U.
	Christian Kaufmann*	U. Zurich
2005	Wesley Rubio	San Diego State U.
	Whitney Qualls*	Auburn University
	Rebecca Trout*	University of Kentucky
2006	Robert D. Anderson	University of Winnipeg
	Linda O'Connor**	University of Delaware
	Joshua R. Ogawa*	Oregon State University
	Matthew Eaton*	Concordia College
	Linda M. Styer*	U. California, Davis
2007	Jennifer Armistead	University of Florida
	Robert D. Anderson*	University of Delaware
	Thomas M. Mascari*	Louisiana State U.
2008	Jerome Schleier	Montana State University
	Christopher Barker*	U. California, Davis
	Lisa Reimer*	U. California, Davis
2009	Alexandra	University of Florida
	Stephanie Larick*	University of Florida
2010	Sarah Wheeler	University of California,
	Kimmy Mains*	University of Kentucky
	Holly Tuten*	Clemson University
2011	Logan Minter	University of Kentucky
	Kristen Meckel-	San Diego County Vector
2012	Jerome Schleier	Montana State University
	Elizabeth Andrews*	University of Kentucky
	Jennifer Gordon*	University of Kentucky
	Joseph Iberg*	University of Georgia
2013	Brian Johnson	Rutgers University
	Andrea Egizi	Rutgers University
	Brittany Nelms	U. California, Davis - CVEC
2014	James Ricci**	University of California
	Eva Bickner***	University of Florida
	Allison Gardner***	U of IL Urbana - Champaign

* $500 cash award presented to winner ** Gerald Hollandsworth Prize *** Honorable mention

AMCA OFFICERS, EXECUTIVE DIRECTORS AND EDITORS

AMCA PRESIDENTS

1935-1939	Thomas J. Headlee*	1968-1969	Thomas D. Mulhern	1995-1996	John D. Edman
1939-1940	Christian T. Williams*	1969-1970	George T. Carmichael	1996-1997	Robert J. Novak
1940-1942	Louis A. Stearns*	1970-1971	Albert W. Buzicky	1997-1998	Gary G. Clark
1942-1944	Robert C. Botsford*	1971-1972	Andrew J. Rogers	1998-1999	Dan L. Ariaz
1944-1945	Robert L. Vannote	1972-1973	Glen C. Collett	1999-2000	William J. Zawicki
1945-1946	Perry W. Ruth	1973-1974	Kenneth L. Knight	2000-2001	David A. Dame
1946-1947	Harry H. Stage	1974-1975	Robert M. Altman	2001-2002	Sammie L. Dickson
1947-1949	H. Duke Peters	1975-1976	Harold C. Chapman	2002-2003	David A. Brown
1949-1950	Harold F. Gray	1976-1977	D. Bruce Francy	2003-2004	Fred W. Knapp
1950-1951	Lester W. Smith	1977-1978	Lewis T. Nielsen	2004-2005	Roger S. Nasci
1951-1952	Don M. Rees	1978-1979	Paul J. Hunt	2005-2006	William R. Opp
1952-1953	Cecil R. Twinn	1979-1980	Glen M. Stokes	2006-2007	Joseph F. Sanzone
1953-1954	Fred C. Bishopp	1980-1981	Robert K. Washino	2007-2008	Gene R. Payne
1954-1955	Roland E. Dorer	1981-1982	Claude H. Schmidt	2008-2009	Major S. Dhillon
1955-1956	Richard F. Peters	1982-1983	Richard C. Axtell	2009-2010	Doug Carlson
1956-1957	Fred L. Stutz	1983-1984	Jimmy K. Olson	2010-2011	Janet McAllister
1957-1958	Arthur W. Lindquist	1984-1985	Gilbert L. Challet	2011-2012	William H. Meredith
1958-1959	John M. Hirst	1985-1986	T. Oscar Fultz	2012-2013	Thomas R. Wilmot
1959-1960	Archie D. Hess	1986-1987	Donald J. Sutherland	2013-2014	Roxanne Connelly
1960-1961	Daniel M. Jobbins	1987-1988	George B. Craig, Jr.	2014-2015	Steve Mulligan
1961-1962	William E. Bickley	1988-1989	Bruce F. Eldridge		
1962-1963	Arthur W. Geib	1989-1990	Judy A. Hansen		
1963-1964	Don W. Micks	1990-1991	Robert C. Sjogren		
1964-1965	John A. Mulrennan,	1991-1992	Matthew Yates		
1965-1966	Anthony W. A. Brown	1992-1993	Cyrus R. Lesser		
1966-1967	Jay E. Graham	1993-1994	John A. Mulrennan, Jr.		
1967-1968	Harry D. Pratt	1994-1995	Chester G. Moore		

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AMCA TREASURERS

1935-1943	Thomas D. Mulhern *	1994-2000	Charles T. Palmisano
1944-1950	Thomas D. Mulhern	2000-2011	Allan D. Inman
1950-1953	Roland E. Dorer	2011-present	Gary Hatch
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		

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SECRETARY, EXECUTIVE SECRETARY, EXECUTIVE DIRECTOR

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

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BUSINESS MANAGER

1995-1999	Pamela D. Toups
1999-2000	Marlene Comeaux
2000-2001	Robertamarie Kiley
2001-2004	Martin. S. Chomsky
2004-2006	Sarah B. Gazi

TECHNICAL ADVISOR

2000-present	Joseph M. Conlon

EDITORS OF *JOURNAL OF AMCA**

1941	Edited by the Publications Committee, Lester W. Smith, Chair†
1942-1943	Edited by the Publications Committee, Ralph W. Vanderwerker, Chair‡
1944	Edited by the Publications Committee, J. T. Hart, Chair
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
2004-2006	Kenneth J. Linthicum
2007-	Lal S. Mian

* - *Mosquito News* became the *Journal* of AMCA in 1985

† - Publication of the Eastern Association of Mosquito Control Workers

‡ - Volume 4, Number 1, was edited by the Publications Committee; subsequent volumes had a single editor

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†	Thomas J. Zavortink, editor, & Lewis T. Nielsen, editor emeritus

* - Prior to 1973 *Mosquito Systematics* was named *Mosquito Systematics Newsletter*

† - In 1995 this publication was discontinued

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