Enhanced Mosquito and Vector Surveillance Services
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List of Abbreviations

Arbovirus – Arthropod-borne virus; virus that is transmitted by insects and other arthropods  
CDC – Centers for Disease Control and Prevention  
CVMVCD – Coachella Valley Mosquito and Vector Control District  
CVEC – Center for Vectorborne Diseases, located at the University of California Davis  
ELISA – Enzyme-linked immunosorbent assay - diagnostic tool for detection of antibodies  
IDF – Intermediate Distribution Frame  
IT Department - Information Technology Department  
MDF - Main Distribution Frame  
NAFTA – North American Free Trade Agreement  
RAMP – Rapid Analytical Measurement Platform – test use to detect presence of WN virus in mosquitoes  
RIFA – Red Imported Fire Ants  
RT-PCR – Real Time Polymerase Chain Reaction – sensitive test for detection of viruses in mosquitoes  
SIQVC- Surveillance Integrated Vector and Quality Control Department  
SLE – Saint Louis encephalitis  
UCD – University of California Davis  
ULV – Ultra Low Volume application - type of control measures to control adult mosquitoes  
WEE – Western equine encephalomyelitis  
WNV – West Nile virus
EXECUTIVE SUMMARY OF NEEDS ASSESSMENT FOR ENHANCED MOSQUITO AND VECTOR CONTROL SERVICES

THE DISTRICT

The Coachella Valley Mosquito and Vector Control District (District) is a special district, which operates under the California Health and Safety Code 2000-2093. The District’s Mission is to enhance the quality of life for the community by providing effective and environmentally sound vector control and disease prevention programs through research, development, and public awareness. The District encompasses 2400 square miles and includes nine (9) cities and unincorporated areas of the Coachella Valley. The District is governed by eleven (11) Trustees, each appointed by one (1) of the nine cities in the Valley and two (2) by the County at Large.

VECTOR SURVEILLANCE AND CONTROL PROGRAM

Informative and rapid surveillance data is the key to an effective response in preventing human and animal diseases associated with currently present or potentially introduced arboviruses in the Valley. If increasing levels of virus activity are detected in the mosquitoes or other surveillance systems, the District response must be immediate and effective to protect the residents of the Coachella Valley.

At the present time, the District has in place an area-wide surveillance program for adult mosquitoes and is capable of limited, in-house testing only for the presence of West Nile virus in mosquitoes. Most of the mosquito samples are sent to an outside agency for testing for the presence of arboviruses.

THE FEASIBILITY STUDY OF THE CURRENT LABORATORY

In 2007, the District started financial planning for future growth to accommodate the need for quick and accurate surveillance data, with the purpose to support more effective mosquito control;

- Through strategic planning and series of discussions and workshops beginning in April 2011, the Board of Trustees decided at their July meeting to hire an architectural firm to conduct a feasibility study of the existing laboratory space;
- The purpose of the feasibility study was to address existing laboratory space and the potential for design upgrade and expansion of available space, with an ultimate goal of providing enhanced, molecular surveillance and control programs;
- The District hired HDR, Architecture, Inc. and the final document/report was presented at the January 2012 Board Meeting;
- The HDR Feasibility study proposed a conceptual plan, which principally grouped the District vector and diseases surveillance and quality control program into three zones, all within the bio-control building, including an enhanced laboratory, support services, and offices.
- In addition, the feasibility study indicated that with the upgrade of the bio-control building and the grouping of surveillance and quality control programs in one building, the vacant space, previously used for surveillance program, would provide much needed space for IT Department and an option for expansion of the men’s locker room, when needed.
The Need Assessment to Enhance Current District Services

The purpose of this study is to identify current and future District needs and “gaps” in the field of vector and vector borne disease surveillance to provide enhanced, effective, and environmentally sound mosquito control and disease prevention services to its constituents.

- **Strengths**
  - The District has professional and technical staff in place to provide rapid molecular surveillance services to the residents of the Coachella Valley.
  - The District has the financial ability to support the capital project of upgrading current facility to provide enhanced molecular testing services to the residents of the Coachella Valley.

- **Weakness**
  - The District relies on an outside agency for most of our arbovirus testing. This causes a delay in our response time to arbovirus threats.
  - The District’s current in-house evaluation of vectors for detection of pathogens causing vector-borne diseases is not delivering sensitive and timely surveillance data which delays response time, and addresses only West Nile virus.
  - The current testing prices are expensive and incur additional shipping costs to an outside facility.

- **Opportunities**
  - The District has in place the Feasibility Study Report that indicates that the current bio-control building has the potential to be upgraded to provide enhanced surveillance of a broad range of vector borne pathogens that cause human and animal disease. This enhanced surveillance data will result in timely public outreach and vector control measures.
  - An enhanced facility will result in cost savings in controlling arbovirus transmission and in weekly arbovirus testing.
  - Reducing the time between virus detection and adult mosquito control operations will result in reduced pesticide use, leading to increased savings in cost of chemicals, cost of labor, and reduced environmental risk.
  - An enhanced facility will improve the efficiency of the Operations and Research Staff.
  - An enhanced facility will provide much needed vacant space, previously used by surveillance program, for expansion of the Information Technology (IT) Department

- **Threats**
  - A longer delay to implement control measures results in a higher risk factor for arbovirus transmission and a higher potential for an increased number of human cases.
  - The District has no control over an outside testing agency, its funding or its reporting time.
  - The current outside testing agency is subject to the State of California budget.
  - The current outside testing agency is subject to the Laboratory Director’s research interests.
  - The function of the current testing agency is not mandated by the State of California.

Improved Response Time

With an upgraded facility, the turnaround time from when mosquitoes are collected in the field to when the District receives arbovirus test results would be decreased. This allows for the response time from mosquito
trapping to adult mosquito control to be decreased by 2-6 days compared to the current arbovirus testing procedure. A likely scenario of this timesaving is illustrated below:

**Typical Response Time Scenarios**

**Current Surveillance**

1. Mon
2. Tue
3. Wed
4. Thu
5. Fri
6. Sat
7. Sun
8. Mon
9. Tue
10. Wed
11. Thu
12. Fri
13. Sat
14. Sun
15. Mon
16. Tue

- Traps set
- Results from CVEC
- Additional traps set to determine spread of virus
- Results from CVEC
- 5-day ULV treatment begins

**Upgraded Surveillance**

1. Mon
2. Tue
3. Wed
4. Thu
5. Fri
6. Sat
7. Sun
8. Mon
9. Tue
10. Wed

- Traps set
- In house results
- Additional traps set to determine spread of virus
- In house results
- 5-day ULV treatment begins

**Legend**
- Initial surveillance
- Expanded surveillance
- Posting of spray route
- ULV treatment

Decreasing the amount of time needed to find arbovirus threats and respond to them decreases the spread of viruses from their point of origin and increases the chances of the District successfully interrupting virus transmission. Improving the timeline of this detection process also decreases the potential for human infection.

**Workflow**

Having a single work and office space for the staff in the surveillance and integrated vector and quality control groups would facilitate collaborations between the groups and streamline some of the experimental procedures. This will increase the productivity of the professional group, allowing them to focus more on developing procedures that will benefit the operations department and overall developing a timely and effective response in situations when the virus is detected in the Valley.

**Additional Benefits**

- Additional Testing Capability
  - Exotic and Emerging Disease Surveillance
  - Insecticide Resistance Testing of control products
  - Red Imported Fire Ant Colony Type Determination
  - Insect Identification
• Cost for arbovirus testing to decrease.
• Already have necessary staff to perform new arbovirus molecular tests.
• Provide space, equipment and staff in a case of other public health state or federal emergencies.

**IMPORTANCE OF MOSQUITO BORNE DISEASE SURVEILLANCE**

The Coachella Valley with its large tourist industry, presence of several North American Free Trade Agreement (NAFTA) trade routes, and location within the migratory bird Pacific flyway is an important point of entry for several mosquito borne diseases and mosquitoes (Lothrop et al. 1995, Meyer et al. 1988). University of California Davis (UCD) researchers have described the introduction and amplification of new forms of mosquito borne viruses such as St. Louis encephalitis (SLE) virus into California, western equine encephalomyelitis (WEE) and documented the invasion of California by West Nile virus (WNV) through Imperial and Coachella Valleys (Reisen et al. 2002). *These findings underscore the importance of District’s current and future mosquito borne disease surveillance program.*

Information derived from the District’s mosquito borne disease surveillance program is used to predict and help prevent epidemics of mosquito borne viruses in the Coachella Valley. The District staff utilizes information regarding the level and location of infected mosquitoes in the Valley to direct preventive and efficient mosquito control and public awareness campaigns. Timely use of information regarding the level of mosquito borne disease activity is crucial. Delay in control measures and further prevention of virus transmission can severely increase amplification of the virus and the risk of human illness and death from mosquito borne viruses (Moore et. al. 1993).
CURRENT CVMVCD MOSQUITO BORNE DISEASE SURVEILLANCE PROGRAM

The District’s current *arthropod-borne virus* (arbovirus) surveillance has two primary components: *mosquito surveillance* and sentinel *chicken surveillance*. Both of these programs are run year round, but with decreased intensity during the winter months of December, January, and February. During these months the temperature is too cold to permit efficient replication of arboviruses in their vectors. Most arboviruses don’t replicate below temperatures of 57°F, and the virus needs to pass through a mosquito vector in order to be transmitted. The cold temperatures of these months also reduce the mosquito populations to low levels. *Transmission cycles between the host and vector during the warm weather here in the desert may take only 7-10 days, and operationally it is important to have the option to detect the virus in the vectors as soon as possible to start control efforts.*

From the months of March through November, the District utilizes 60 modified Center for Disease Control (CDC) CO₂ traps and 25 gravid traps. These trap numbers change slightly year to year in response to habitat changes in the Valley. Both trap types are used to collect mosquitoes from the field, but they differ on what mosquitoes they attract and capture effectively. In the Coachella Valley, there are 12 mosquito species detected; however currently, only *Culex tarsalis*, the encephalitis mosquito, and *Culex quinquefasciatus*, the southern house mosquito, are tested for WNV, SLE and WEE.

**CDC CO₂ baited traps**

- Good at catching a broad range of species of *host-seeking* mosquitoes. These mosquitoes can be seeking their first blood meal or it can be a second, third or possibly later blood meal.
- Good at catching *Culex tarsalis* mosquitoes which are the primary rural vector of WNV, as well as, WEE and SLE.
- Less effective at catching of *Cx. quinquefasciatus*, the most important urban vector of SLE, WEE and WNV in the Coachella Valley.

**Gravid Traps**

- Good at catching *Cx. quinquefasciatus*
- Designed to capture mosquitoes that have already taken a blood meal and searching for a water source to deposit their eggs.
- Less effective at catching many *Cx. tarsalis* mosquitoes.

**Mosquito Samples in the Lab**

- Mosquitoes are transported to the District surveillance lab first thing in the morning after the traps were set the prior afternoon.
- It is important that the mosquitoes be brought back *alive*.
- Many arboviruses including WEE, SLE and WNV are not very stable outside of living organisms and quickly degrade unless frozen around - 80 °C, making virus detection nearly impossible.
Mosquito samples are anesthetized.
Once the mosquitoes are immobilized they are sorted to the species level and counted.
From each trap, up to 5 mosquito samples, of each species, with at least 5 and up to 50 mosquitoes in each sample, are separated, labeled and held at – 80 °C environment until shipping.

**Shipping of Mosquito Pools**

**TRAPPING ON MONDAY** *(half of total number of traps set-up timeline)*

- Traps collected on **Tuesday** and processed **the same day**;
  - Mosquito samples are shipped overnight on dry ice to the Center for Vectorborne Diseases (CVEC) **the same day** *(Tuesday)*.
  - **Results** received from CVEC by **Thursday; 4 days after trapping**.
  - **Operations Department** starts organizing control efforts **4 days** after trapping.
- Traps collected on **Tuesday, but not** processed the same day because of the other duties that delayed counting, sorting of mosquitoes at the District, **OR** testing at CVEC delayed.
  - Mosquito samples processed **the next day** *(Wednesday)* and sent overnight same day *(Wednesday)* on dry ice to CVEC **OR** tested at CVEC delayed.
  - **Results** received by **Friday; 5 days after trapping**.
  - **Operations Department** starts organizing control efforts **7 days** after trapping (normal or emergency planning).

  * If Monday is a holiday then all of this is delayed by one day.

**TRAPPING WEDNESDAY** *(other half of total number of traps set-up timeline – mostly gravid traps and CDC CO₂ traps placed by sentinel chickens)*

- Traps collected on **Thursday** and processed the same day;
  - Mosquito samples are shipped overnight on dry ice to CVEC the following **Monday**.
  - **Results** received from CVEC by **Wednesday; 7 days after trapping**
  - **Operations Department** starts organizing control efforts **8 days** after trapping.
- Traps collected on **Thursday, but not** processed the same day because of the other duties that delayed counting, sorting of mosquitoes at the District, **OR** testing at CVEC delayed.
  - Mosquito samples processed **the next day** *(Friday)* and sent overnight **next Monday** on dry ice to CVEC
  - **Results** received by **Wednesday; 7 days after trapping**.
  - **Operations Department** starts organizing control efforts **8 days** after trapping.

**Sentinel Chicken Surveillance**

During the weeks when lab staff does not place CO₂ traps on Monday, they are conducting arbovirus surveillance by taking blood samples from the chickens at the sentinel chicken flocks. The District has 13 flocks of 10 chickens each. The results of the sentinel chicken samples are a later indicator of arbovirus transmission than positive mosquito collections. The reason for this is that the chickens must become infected by the bite of an infected mosquito and then develop an antibody response large enough to be detected by Enzyme-linked
immunosorbent assay (ELISA) testing. The benefit to this testing is that the chickens are in the field 24 hours a day, 7 days a week, and thus can pick up transmission that mosquito collecting might miss due to a factor affecting mosquito trapping on a particular night, i.e. extreme wind, rain, extreme temperatures, etc. Sentinel chickens can also be used to estimate mosquito to human virus transmission potential better than positive mosquito collections.

The blood samples from each of these chickens are tested for the presence of antibodies to SLE, WEE, and WNV. The District currently sends the samples to the California Department of Public Health for testing but is working on performing these tests in house. The District already has the needed equipment to perform ELISA, the test used to determine if the chicken blood samples have antibodies to the previously mentioned arboviruses.

*One of the issues that the laboratory staff experiences currently when performing ELISA testing, is that the work flow is disrupted, since the equipment needed for testing is located in two labs and requires staff to go from one building to the other to accomplish the task.*

**NEEDS FOR DISTRICT MOSQUITO BORNE DISEASE SURVEILLANCE AND SOLUTIONS**

Major benefits to having an upgraded lab that can perform molecular, genetic-based testing:

*Faster turnaround times with arbovirus surveillance results and timely mosquito control effort*

1. When performed on site these tests can be completed the same day that the mosquitoes are received.
2. If the test cannot be completed the same day then arbovirus test results can easily be completed the day after mosquitoes are collected.
3. *This rapid analysis is especially beneficial when there have been positive virus results and the lab staff expands the number of traps around a positive location to try and determine how widespread the arbovirus virus may be and where would be the most important areas for operations to conduct mosquito control efforts.*
4. The District staff can reorganize work hours and perform testing evenings and weekends if needed, and the traps collected Thursdays and Fridays can have results back the same day rather than a 6-7 day wait due to the weekend and delays between testing and shipping.

The time difference between the rapid in-house testing versus relying on sending mosquitoes to CVEC for arbovirus detection can have a significant impact on when operations starts intensified larviciding and adulticiding to control adult populations of vectors in an area with WNV (or another virus) transmission. Delay in response and spread of arbovirus will also result in higher costs associated with mosquito control efforts.

**OPERATIONAL RESPONSE AFTER DETECTING VIRUS IN THE AREA**

The District gets preliminary positive test results from using Rapid Analytical Measurement Platform test (RAMP), a less sensitive test than RT-PCR test used by CVEC. The reason that the District waits until the
surveillance data is verified by CVEC is that initiating a full response to an arbovirus threat includes a number of steps such as:

- press releases,
- determining spray routes,
- alerting the Agriculture Commissioner’s office of our spray route,
- posting spray routes on the District website as well as in the neighborhood where the adulticiding will take place,
- running the spray route for at least 5 consecutive nights (weather permitting)

All of these listed steps are expensive and labor-intensive. The District’s goal is to minimize adult mosquito control since the control agents used for suppression of adult mosquito abundance could have higher risk of developing resistance if used too often. Adulticides may have more negative environmental impacts when compared with controlling mosquito larvae. In addition, control of adult mosquitoes is not as effective as controlling mosquito larvae. Because of these reasons, the District only targets adult mosquitoes for control efforts when the potential of interrupting active virus transmission is high. *The only way to know when this is possible is when there is reliable surveillance data that delineates where the virus is located in the Valley, when it was detected, and how many and what species of mosquito is transmitting the virus.*

**OPERATIONAL RESPONSE TIMELINE**

**DETECTION OF POSITIVE MOSQUITO SAMPLE**

How many days a full operational response is delayed due to waiting for surveillance results is illustrated in an example in the section below, *taken from WNV transmission season detected in August 2011 in Cathedral City.*

- With the District’s current capabilities, the time of initial virus detection to a full operational response in an area of virus transmission is between **5 to 12 days**. This depends on a number of factors including the number of WNV positive samples initially detected, when the samples were collected, how conclusive results from using in-house test, and if testing on site is used.
- If RT-PCR testing were available to be performed at the District then the time from initial virus detection to a full response from the District to control virus transmission in an area would be **3 to 5 days**.
- *Transmission cycles between the host and vector during the warm weather here in the desert, may take only 7-10 days, and operationally it is important to have option to detect the virus in the vectors as soon as possible and start control efforts as soon as possible.*
- Reducing the time by 2 to 7 days to begin controlling adult mosquitoes can have a profound effect on the spread of arbovirus transmission. Research conducted on vector mosquito species found in the Coachella Valley show that the vectors travel on average 1 km in 2 days and 2 km in 3 days (Reisen and Lothrop, 1995). Some mosquitoes in the Valley have been known to travel much further than this. Allowing mosquitoes to travel for 2 to 7 extra days substantially increases how far a mosquito-borne
pathogen may travel and increases how much more additional surveillance and control may need to be required to halt virus transmission.

- The longer mosquitoes carry the virus the more chances there are for people to get infected with the virus vectored by the mosquitoes.
- Additional time also may mean that the mosquitoes fly further from the initial site of infection, leading a larger area that needs to be covered, increased use of adult mosquito control measures, and increased labor and chemical costs.

There are still many variables that affect how soon the District will respond by an adulticiding campaign such as how many samples of WNV were found positive when pools of mosquitoes are first tested, and if mosquitoes required more than one day for sorting and counting.

**CASE STUDY: CATHEDRAL CITY WNV SURVEILLANCE AND RESPONSE AUGUST 2011**

Mosquito borne disease surveillance that began on *Wednesday, August 3rd 2011* led to an intensified month-long mosquito surveillance and control effort in Cathedral City. District staff carried out two - 5 day ULV adult mosquito control missions in an effort to prevent disease transmission from infected mosquitoes within the WNV focus area.

The level of WNV positive mosquito samples and where they occur in space and time influences how District staff utilizes its Emergency Response Plan, surveillance tools, and treatment strategies. The flow charts that are depicted below represent District surveillance and control operations that occurred in Cathedral City in August of 2011. Also depicted is one possible scenario that would be feasible if the proposed Integrated Vector Control Lab is upgraded. *Remember timing of public outreach and mosquito control strategies is integral to the prevention of mosquito borne disease amplification and transmission to humans.*
Figures 1 and 2: Maps depict timing of surveillance, reporting and spread of West Nile Virus (WNV) in Cathedral City during August 2011. The red dots represent traps with mosquitoes found to be positive for WNV and white dots represent traps with no mosquitoes positive for WNV. The red line represents the expected range of infection of mosquitoes by WNV.
August 2011 – Actual Cathedral City Surveillance and Control

In this instance, the WNV positive mosquito sample was from a gravid trap that was set on a Wednesday and collected on Thursday. Thus, the sample was not shipped for testing until the following Tuesday with other samples collected on that Tuesday in CO₂ traps, as is standard operating procedure. Typically, WNV positive samples are first detected in the rural areas, but this was the first WNV positive sample of the season and was found in a city. This unusual combination of events meant that we wanted to verify that WNV transmission was occurring where the sample was collected before proceeding with adult mosquitoes control efforts.

Hypothetical Surveillance and Control Upgraded Lab – Emergency Planning

In this hypothetical scenario, the WNV positive mosquito sample was detected on Day 8 after the trap was set on Day 2. The sample was tested on Day 9 and found to be positive for WNV. On Day 11, the sample was tested again and found to be positive for multiple WNV positives. The ULV mission to control adult mosquitoes was conducted on Days 6-10. The ULV mission to control larval control was conducted on Days 15-19.
TESTING COSTS

In order to perform RT-PCR testing, specific equipment and reagents are needed for the extraction and amplification of the genetic material.

- The cost of equipment to perform these tests is about $70,000. Because these instruments require a high level of precision to perform, and because of the cost of fixing them if broken is expensive, an annual service contract should be purchased as well.
- The annual service contract for the necessary equipment is approximately $6,700/year.
- There are also disposable reagents that are used for each test which would cost about $6 per test performed.

CURRENT COST

MOSQUITO SAMPLES TESTING

- Cost of testing mosquitoes is $20 per sample with CVEC.
- District is sending about 3,000 x $20 samples/year = $60,000.
- The District doesn’t currently pay that amount because of research projects that are involved in with Dr. Reisen, who covers the cost of testing over 1,500 samples.
- The District current cost - 1,500 x $20 = $30,000/year
- When our research agreements with Dr. Reisen end, we will need to cover the cost of all (3,000) of the mosquitoes samples - $ 60,000.
- The Districts sends an average of 17 overnight shipments of mosquitoes to CVEC for testing at $100 each
- The District sends an average of 18 standard 2-day shipments of mosquitoes to CVEC for testing at $25 each
- We pay an average of (17 x $100) + (18 x$25) = $2,150/year for shipping samples for testing

FUTURE COST

MOSQUITO SAMPLES TESTING

- Cost of testing mosquitoes would be $6.00 per sample
- The District cost for testing the same amount of samples - 3,000 x $6.00 = $18,000.
- The District will save $37,000 per year ($60,000 CVEC testing cost + $2,150 shipping cost) - ($18,000 testing cost + $6,700 maintenance cost) ~ $37,000.
- The District will pay off the equipment in 2 years.
ADDITIONAL BENEFITS

EMERGING/EXOTIC VIRUS DETECTION

In addition to more rapid virus detection, being able to perform RT-PCR at the District would have other benefits. One of these benefits is that the District would be able to perform testing for arboviruses if CVEC stopped performing this service for mosquito control districts. It was announced at the Mosquito and Vector Control Association of California (MVCAC) meeting in Burlingame, CA held from Jan 29th – Feb 1st, 2012 that the CDC had reduced supporting states with arbovirus testing including WNV surveillance. This was not a surprise as the CDC has put arbovirus support up for major reductions as the budget cuts to the CDC have been proposed. The state budget is also cutting funding to the UC system. If these trends continue there could be an end of virus testing performed on mosquito pools for districts.

Virus surveillance in mosquito pools can also be used as an early detection method for emerging or exotic pathogens moving into a new area. If such an arbovirus is detected early in mosquitoes then the virus transmission can be eradicated, human health may be preserved, and money can be saved from trying to control a widespread outbreak. A molecular based assay like RT-PCR has the capability to test for any virus, and costs for supplies needed to test for new viruses would be very low.

California has had a long history of international travel and commerce. With the new methods of rapid global transportation, people, animals, and cargo are moved from various parts of the globe with great speed into the state. This allows for vectors and pathogens to be more easily transported into the state now than at any other time in our history.

The Global Air Network of the 21st century.
In 2011, *Aedes albopictus*, the Asian tiger mosquito, was found in the Los Angeles suburb of El Monte. This extremely invasive mosquito from Southeast Asia has already invaded most of the Eastern U.S., parts of Europe, Central and South America and some countries in Africa. This mosquito poses a public health threat as it is a vector of important human pathogens such as dengue and chikungunya. Some of the arthropod-borne diseases that have been recently invasive in other parts of the world include Venezuelan Equine Encephalitis Virus in Latin America, Japanese Encephalitis Virus in Southeast Asia, Rift Valley Fever in Africa and the Middle East and chikungunya in Europe. There are competent vectors for all of these viruses currently in California and the Coachella Valley except for chikungunya which is transmitted by *Ae. albopictus*, detected but not fully established.

**GENETIC PESTICIDE TESTING**

One of the most important aspects of vector biology that a mosquito control district needs to know is if the mosquito populations that they are trying to control are resistant to any of the control methods that they are using. If the mosquitoes are not killed by the products that are being used to control them then the district is wasting time and money, not controlling the vectors and likely using more pesticide than they normally would as control efforts are ineffective.

The most common way to determine if there is insecticide resistance in a population of mosquitoes is to rear a control colony of mosquitoes that is never exposed to pesticides, and then bring in mosquitoes from the field. These mosquitoes are then exposed to the same doses of pesticides and differences in survivorship are measured. If the wild-caught populations show a statistically significant difference in having more of their population survive the exposure to pesticide compared with the laboratory reared population of mosquitoes, then resistance has been developed in natural/field populations of mosquitoes.

Map showing where *Ae. albopictus* was discovered in 2011 in the LA area

The different colored polygons represent different locations where surveillance for the mosquitoes was performed. The yellow push pins represent locations where the invasive mosquito was found.
Some of the benefits to these tests, referred to as bioassays, are that they don’t require any special equipment and they are cheap to perform. On the downside, mosquitoes from the field must be reared to the right developmental stage to use in tests, so that the ages of the mosquitoes in the wild and lab populations are the same. This can be a problem as mosquitoes from the field do not always take blood meals, reproduce and lay eggs in a cage. This is especially true of *Cx. tarsalis*. In addition, pesticide resistance can be a localized process, so mosquitoes from different parts of the Valley must be tested to determine where resistance is present.

*Currently, the District does not have adequate rearing rooms for mosquitoes and that was discussed in more detail in the Feasibility Study, submitted by HDR in January 2012. The proposed upgrade of the facility will provide adequate space and condition for rearing mosquitoes and performing testing to detect potential of resistance of wild mosquitoes.*

**CENTRALIZATION OF SIQVC STAFF AND IMPROVING WORKFLOW**

The division of the equipment between the labs in the Operations and Biocontrol Building does not allow surveillance and research tasks to be conducted in a productive and efficient manner.

The Surveillance, Integrated Vector and Quality Control department (SIQVC) staff is currently split into three locations. The surveillance lab group is all located in the Operational Building with the offices of the Environmental Biologist, Operations Biologist and Fishery Biologist located in one of the operations rooms, out of the surveillance lab while, the Biologist and the Tank Room Maintenance worker are based in the Biocontrol Building.

This separation of the staff is not as conducive to frequent communication as it would be if the whole department was housed in one area. It also makes constant oversight by the supervisors tedious to perform, especially for the Environmental Biologist who must walk from the Operations Building over to the Biocontrol Building and the Tank Room to observe, communicate and work with assigned staff. Also there are labs in the Operations Building and the Biocontrol building which house different pieces of necessary equipment to perform certain tests, so the staff must carry lab samples and supplies back and forth between the two labs to carry out certain experiments. If there was a central lab space to house all the laboratory equipment and supplies it would make performing certain tasks, such as sentinel bird serum assays for arbovirus exposure (ELISA) easier and safer for the lab staff.

**RED IMPORTRED FIRE ANT (RIFA) REARING /QUALITY CONTROL**

A large percentage of the District’s services to the Coachella Valley is the surveillance and control of red imported fire ants (RIFA). Currently, the District does not have the appropriate space to house a colony of RIFA for in-house testing of surveillance and control products. Construction of the new surveillance lab would provide ample space to rear and study these organisms which would increase the ability of District Staff to assess the efficiency and effectiveness of our RIFA Control

**GENETIC ANALYSIS OF RIFA COLONIES**

Our staff is working with researchers from Texas A&M to map the colony type (monogyne or polygyne) of RIFA across the Coachella Valley. This has implications as to how effective control methods are currently, as well as determining how different treatment strategies in the Valley may work in the future. Areas populated with monogyne (single queen) colonies will have lower ant densities, and may have been under control pressure in the past. These colonies can also require more extensive baiting to eliminate colonies, as the ant population is
made of more numerous, but smaller colonies, that won’t share resources like polygyne (multiple queen) colonies will. Monogyne colonies are also much less susceptible to the natural red imported fire ant pathogen *Kneallhazia*. This microsporida has been show to help directly and indirectly control RIFA colonies and was recently discovered in fire ants located in the Coachella Valley.

Tracking the changes in colony distribution of RIFA could allow the District to help gauge the success of the program as well as determine what control strategies to employ in different regions within the District. Currently we test for colony type and the presence of *Kneallhazia* by sending ants to a lab in Texas. Our contract for doing this expires at the end of the fiscal year, and it is $100 per ant sample for the testing. These tests could be performed in-house at about 1/10 of that rate.

**REUSE PLAN FOR OPERATIONS BUILDING LABORATORY**

The existing Laboratory Office Space in the Operations Building is to be utilized by the Information Technology (IT) Department. These rooms include:

- Research Office (RO)
- Vector Ecologist Office (VCO)
- Vector Biologist Office (AVE)

Centralization of the Information Technology Department in the existing Laboratory Office space provides end-users and members of the IT Department one area to communicate and collaborate on issues affecting software end-users.

The Research Office provides two options for the District:

1. The office can serve as the District’s Secondary IDF (Intermediate Distribution Frame), a cable rack that interconnects and manages the telecommunication wiring between MDF (Main Distribution Frame) and workstation devices. The office provides the District 360 degrees of working space around the IDF allowing enough room to move and reconfigure devices as needed. The installation of a proper distribution system for conditioned cold air would will be possible, as well as, pursuing a Waterless Fire Suppression System to protect the District’s Information Technology investments.

2. The office can also serve as a drafting room for employee’s to utilize the 42” inch plotter, a computer printer, and the 42” inch cutter in the production of large scale banners, posters and maps. Printer supplies (toners, fusers, plotter rolls, and paper), replacement devices, and new devices would be stored, inventoried and monitored by the Information Technology Department.

The Vector Ecologist Office would be reassigned to accommodate the District’s Geographic Information Systems Analyst, while the Vector Biologist office would be reassigned to accommodate the Assistant Information Technology/Geographic Information System Personnel.
**Information Technology Office**
The men's locker room will be expanded out into the current Information Technology Office allowing for additional lockers, showers and stalls. The newly expanded dressing room provides adequate space to accommodate both divisions of Operations (Mosquito & Red Imported Fire Ant), as well as the District’s Seasonal Staff at the level of 5-6 seasonal staff per shift. Any increase in number will create “traffic jam”. A properly constructed “mud room”, a secondary entryway to remove and store footwear, eliminates Vector Control Technicians tracking in mud into the men’s locker room and Operations Office Space.

**REGIONAL TESTING CENTER**
Currently there are seven vector control districts in California that have their own BSL 2/3 lab to conduct in-house RT-PCR testing on mosquito and tick samples. One District currently tests samples for other Vector Control Districts. If the proposed upgrade of current Biocontrol laboratory is approved, one potential function of the lab could be to serve as a regional mosquito virus testing center for Southern California vector control agencies, if needed. However this would require additional staffing.

**ALTERNATIVES**

**ALTERNATIVE #1 - Keep the “Status Quo”**
Continue to send mosquito samples and sentinel chicken sera samples to CVEC and California Department of Public Health for disease testing.

**Advantages:**
- Not incur costs of upgrading laboratory and associated maintenance.
- No need to change current surveillance and testing protocols.

**Disadvantages:**
- Reliance on external organizations for vitally important disease surveillance information when future of organizations is unstable due to change in leadership and economic instability within the state;
- No flexibility in testing times when needed to direct intensified public outreach and vector control measures to protect public health.
- Potential for increase of service cost for mosquito samples testing in near future.
- Continued costs in shipping and dry ice

**ALTERNATIVE #2 - Testing Performed by other Vector Control District**
Send samples to another Vector Control District currently testing mosquito samples / sentinel chicken sera samples for mosquito borne viruses by RT-PCR

**Advantages:**
- Not incur costs of upgrading laboratory and associated maintenance.
- Testing rates currently cheaper than CVEC rates.
• May be able to perform testing if state and CVEC lose funding.

Disadvantages:

• Reliance on external organizations for vitally important disease surveillance information.
• Only test for WNV.
• Loss of flexibility of testing times when needed seasonally to direct intensified public outreach and vector control measures to protect public health.
• Other mosquito control districts may not be able to accommodate all of our samples, especially if additional districts also send their mosquito samples for testing.
• Potential increase in cost for testing because of increase volume for testing and need for additional labor.
• Potentially change in District surveillance and testing workflow not suitable for the District.
• Continued costs in shipping and dry ice
REFERENCES


