

# Guidance for Surveillance of and Response to Invasive *Aedes* Mosquitoes and Dengue, Chikungunya, and Zika in California

California Department of Public Health

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This document was prepared by the California Department of Public Health, Division of Communicable Disease Control, with input from the Mosquito and Vector Control Association of California and the California Conference of Local Health Officers.

## Objective

This document was developed to guide local vector control agencies and health departments to prepare for, conduct surveillance of, and respond to the detection of invasive *Aedes* mosquitoes and human cases of dengue, chikungunya, Zika, or other exotic *Aedes*-borne viral infections potentially transmitted by these mosquitoes. *Aedes aegypti*, also known as the “yellow fever mosquito,” is the primary vector of these viruses and has established populations in over 400 cities within 24 counties in California. *Aedes albopictus*, also known as the “Asian tiger mosquito,” is also a capable vector currently present in parts of Los Angeles and Shasta counties. Although none of these viruses are endemic in California, local transmission is possible in cities with invasive *Aedes* mosquitoes as travelers return and visitors come from areas with known virus transmission. A comprehensive and collaborative local action plan should be developed to address the detection and establishment of invasive *Aedes* mosquitoes and potential transmission of non-endemic mosquito-borne viral infections.

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*It is recommended that vector control and health department staff read all parts of this document to better understand the activities involved under different scenarios and to coordinate the appropriate activities at the local level.*

## Introduction

The discovery of *Aedes albopictus*, also known as the Asian tiger mosquito, in 2011 in Los Angeles County, and *Ae. aegypti*, also known as the “yellow fever mosquito,” in 2013 in urban areas of Fresno, Madera, and San Mateo counties demonstrated that California was vulnerable to colonization by these highly invasive mosquitoes. By the end of 2024, *Ae. aegypti* had spread to over 400 cities within 24 counties, and *Ae. albopictus* had established populations in some areas of Los Angeles and Shasta counties. Both species are vectors of exotic arthropod-borne viruses (arboviruses) including dengue, chikungunya, Zika, and yellow fever. Every year, travel-associated human cases of *Aedes*-borne viral illness are reported in California. In 2023 the first two locally acquired cases of dengue were reported in Los Angeles County residents, and in 2024 locally acquired cases of dengue were reported in residents of Los Angeles, San Bernardino, and San Diego counties, all in areas where one or both species are established. The source of the virus for local mosquitoes was viremic individuals returned from a dengue-endemic area.

- Dengue is a viral disease characterized by fever, headache, and joint and muscle pain, which can progress to bleeding and shock in some people. Dengue is the most widespread *Aedes*-borne virus with transmission throughout much of the tropics. Localized outbreaks have occurred in areas of the United States where *Ae. aegypti* and *Ae. albopictus* are established, including Arizona, Florida, Hawaii, and Texas.
- Chikungunya is a viral disease characterized by fever, rash, and severe joint pain. Outbreaks have occurred in countries in Africa, Asia, Europe, and the Indian and Pacific Oceans. In late 2013, the first local transmission of chikungunya virus in the Americas was identified in the Caribbean Islands, followed by rapid spread to other countries in South and Central America. Transmission continues to occur in tropical and subtropical regions across the globe.
- Zika is a viral disease characterized by fever, rash, and joint pain. Before 2015, outbreaks had occurred in areas of Africa, Southeast Asia, and the Pacific Islands. In May 2015, human cases were detected for the first time in Brazil, and Zika spread rapidly to other countries throughout the Americas. Zika was initially considered a mild disease, but there is now a clear association between Zika infection during pregnancy and birth defects such as microcephaly — the development of an abnormally small head and brain. In adults, Zika infection has been associated with Guillain-Barré syndrome, an autoimmune neurological disease. Incidence of Zika transmission worldwide has dropped significantly since 2022.
- Yellow fever is a viral disease that can cause illness with symptoms ranging from fever with aches and pains, to life-threatening liver disease with internal bleeding and yellowing skin (jaundice). Although the Americas have a long history with yellow fever, the last U.S. outbreak occurred over a century ago. The virus continues to exist in tropical and subtropical areas of Africa and South America but is a very rare cause of illness in U.S. travelers, in part due to the availability of an effective vaccine, and thus is currently not a focus of this document.

The behavior and habitat preferences of *Ae. aegypti* and *Ae. albopictus* differ substantially from the *Culex* species that are the primary targets of mosquito control programs in California's urban areas. Adult *Ae. aegypti* and *Ae. albopictus* are active during the day, have short flight ranges, and females are aggressive and persistent biters of mammals, especially humans. What is most distinctive is their preference for small, water-holding containers for laying eggs and larval development, hence they are known as "container-breeding" mosquitoes. Their close association with and dependence on humans to provide larval habitats, particularly within residential properties, often results in a widespread but patchy distribution, making effective surveillance and control a challenge. Detection and control are further complicated by eggs that resist desiccation and can remain viable for months on dry surfaces of containers. It can be difficult to determine the origin of *Ae. aegypti* and *Ae. albopictus* infestations, but transport of dormant eggs via imported tires and house plants has been associated with introductions of these mosquitoes into California. Individuals moving materials via planes, ships, cars, or other vehicles from infested areas to non-infested areas also facilitates spread. Once established, these mosquito species are typically very difficult to eliminate from residential areas.

The environmental epidemiology of *Aedes*-borne viruses can be difficult to track due to elusive infections in both mosquitoes and humans. Testing female mosquitoes for virus infection rarely produces positive individuals and a large percentage of human infections are asymptomatic or sub-clinical in nature and thus are never identified or reported. Voluntary blood draws from people living in areas surrounding a known case also rarely identify additional infections; people who are closely associated with known human infections are the most likely to test positive. Monitoring the abundance and infection prevalence of mosquito vectors is useful for anticipating outbreaks of certain arboviruses such as West Nile virus, but in the case of *Aedes*-borne viruses, it is more likely for transmission to be detected first in humans. As a result, transmission risk using environmental indicators is difficult to evaluate, as *Aedes*-borne viruses can circulate at low levels among mosquitoes and humans without being detected.

The prevention or reduction of transmission of *Aedes*-borne viruses is dependent on limiting human-mosquito contact through mosquito control and personal protective measures. It is important that local vector control agencies, health departments, and other agencies work collaboratively to raise public awareness of these mosquitoes and the mosquito-borne viruses they can transmit, as well as develop and implement proactive surveillance and response plans. Early detection and data-driven response are critical to protect public health and minimize the risk of local disease transmission.

## Recommended Actions for Local Agencies

The recommended surveillance and response actions for vector control agencies and health departments will differ based on several factors including the presence or absence of invasive *Aedes* mosquitoes, whether human infections with dengue, chikungunya, or Zika have been identified, and evidence of local virus transmission. Support services available to local agencies by the California Department of Public Health (CDPH) are listed at the end of this section.

## Recommendations for Local Vector Control Agencies

### Pre-Detection of *Aedes aegypti/albopictus*

- Identify local, state, and federal agencies and resources that can be consulted regarding identification, surveillance, and control of *Ae. aegypti* and *Ae. albopictus*.
- In coordination with the local health department, develop a response plan that can be implemented at the first detection of invasive mosquitoes. The plan should include preparedness for enhanced mosquito surveillance and control activities, protocols and responsibilities for sharing information about human cases of dengue, chikungunya, and Zika, working drafts of public education materials, and agreements (including mutual aid), with neighboring health departments and vector control agencies to provide assistance if needed.
- Develop and implement an early detection plan for invasive mosquitoes.
  - Ensure staff can identify all life stages of *Ae. aegypti* and *Ae. albopictus*.
  - Notify CDPH of any mosquitoes tentatively identified as *Ae. aegypti* or *Ae. albopictus*; send specimens or photos to confirm identification if needed.

- Initiate an education and outreach program designed to educate and mobilize the public to report daytime-biting mosquitoes and eliminate larval sources.
- Ensure receptionists are trained to ask appropriate questions to walk-in and call-in customers relative to invasive mosquitoes and recognize when information warrants a precautionary follow-up inspection or referral to the local health department.
- Deploy strategically placed, target-specific egg and adult mosquito surveillance traps. See **Appendices A** and **G**.

### **Initial Detection of *Aedes aegypti*/*albopictus***

- Immediately notify the local health department, CDPH, and neighboring vector control agencies when *Ae. aegypti* or *Ae. albopictus* mosquito identification is confirmed; request assistance for follow-up response if indicated.
- Discuss with CDPH observations and findings of confirmed invasive mosquitoes, potential infestation areas, and possible introduction and movement pathways.
- In coordination with the local health department, distribute public education materials, including a media release, describing the discovery of invasive mosquitoes and the disease risks they present. Reassure the public that human transmission risks are low and request that the public contact the local vector control agency regarding daytime-biting mosquitoes. See media release template, **Appendix B**.
- Enhance mosquito egg and adult surveillance (e.g., ovi- and adult traps) and larval surveillance (e.g., property inspections) to delineate the infested areas.
- Initiate a door-to-door campaign in urban areas surrounding the point(s) of discovery to:
  - Distribute public education materials urging the public to empty or discard small containers of standing water and take personal prevention measures to reduce mosquito bites.
  - Gain permission to conduct larval surveillance on selected residential or commercial properties and, if a desirable location, to place ovi- and adult mosquito traps; educate property owners regarding habitat reduction.
  - Apply Environmental Protection Agency (EPA)-registered products to control immature and adult mosquitoes on private property if necessary.
- When indicated, initiate area-wide control of immature and adult mosquitoes using EPA-registered products.
  - Define areas of control based on surveillance data, including presence, relative abundance, and distribution of invasive *Aedes* within the urban environment.
  - Products can be applied on foot and with vehicle-mounted sprayers. Depending on the extent of the infestation, local topography, and environmental conditions, aerial applications also can be considered, especially if a threat of local arbovirus transmission is suspected.
  - Removal of potential larval habitats and chemical control of adults and larvae on individual properties within the target area are recommended when time and resources permit.

## Travel-Associated or Locally Acquired Human Infection(s) Identified

- If notified by the local health department of any suspected, probable, or confirmed travel-associated or locally acquired case(s) of dengue, chikungunya, or Zika that may have been viremic while being in an invasive *Aedes*-infested area:
  - Request the case-patient's residential address and any additional information on other areas the patient may have visited while potentially viremic.
  - Ensure patient confidentiality by protecting any personal identifiers including name, address, or other personal information.
  - Ensure staff are trained regarding laws that govern the use of confidential information.
- If epidemiological data confirms locally acquired infection(s), work with the local health department and CDPH to issue a joint media release ensuring patient confidentiality. See media release template, [Appendix B](#).
- In coordination with the local health department, immediately implement enhanced adult mosquito surveillance and control around the case-patient's residence (maintaining patient confidentiality) to prevent or interrupt the virus transmission cycle. A target area of 100-200m radius is recommended, but can be increased or decreased in size based on local surveillance data.
- Investigate other locations where the patient may have been exposed to invasive *Aedes* mosquitoes while viremic and implement similar enhanced adult mosquito surveillance and control if indicated.
- Distribute public education materials within the affected area(s) to raise awareness about invasive *Aedes* mosquitoes, the viruses they can transmit, symptoms of disease, and use of personal protective measures.
- If invasive *Aedes* mosquitoes are identified within the target area, initiate area-wide control of immature and adult mosquitoes using EPA-registered products following recommendations outlined in the previous section, **Initial Detection of *Aedes aegypti/albopictus***.
- If resources permit, pools of female mosquitoes ( $\leq 50$  mosquitoes per pool) collected from the affected area(s), or other areas of concern, can be sent to the Davis Arbovirus Research and Training (DART) laboratory for arboviral testing. See [Appendix F](#).
  - The local health department, CDPH, and neighboring vector control agencies should be notified of any infected mosquitoes.
  - The finding of virus-infected mosquitoes is indicative of potential local transmission. Further enhance public outreach and mosquito surveillance and control in and around the area where infected mosquitoes were collected.
  - If virus-infected mosquitoes are found, consider issuing a media release in coordination with CDPH and the local health department to raise awareness of increased virus transmission potential. If appropriate, state that no locally acquired human cases have been confirmed.
- Continue to closely monitor for invasive *Aedes* mosquitoes within the identified area(s) of concern.

- Use local mosquito and human surveillance data and climatic conditions to guide the follow-up timeframe. Implement additional control measures as necessary.
- Typically, three extrinsic viral incubation periods in mosquitoes (approximately 45 days; see [Human Disease Surveillance](#) section) with no additional human cases identified suggests that transmission has been interrupted.
- Continue to encourage the public in reporting daytime-biting mosquitoes, reducing larval habitats on their properties, and taking personal protective measures to prevent mosquito bites.
- Escalate and expand all activities in parallel with increases in the number of human cases identified.

## Recommendations for Local Health Departments

### Pre-Detection of *Aedes aegypti/albopictus*

- Identify local, state, and federal agencies and resources that can be consulted regarding human case surveillance and laboratory confirmation for suspected cases of dengue, chikungunya, and Zika infections.
- In coordination with the local vector control agency, prepare a public education plan that can be implemented at the first detection of invasive *Aedes* mosquitoes. A similar plan should be prepared for the first detection of locally acquired human infections with dengue, chikungunya, or Zika virus. Where no local vector control agencies exist, coordination should be with CDPH. The plans should include a media release and other relevant public education materials.
- Establish with the local vector control agency protocols/procedures for sharing need-to-know case information, allowing for rapid response to control the spread of disease-carrying mosquitoes.
- Continue to report to CDPH via the California Reportable Disease Information Exchange (CalREDIE) or, for non-participating jurisdictions, by fax or secure email, any suspect, probable, or confirmed case of dengue, chikungunya, and Zika virus infection; ensure report includes patient(s) symptom onset date and travel history.
  - If the case-patient(s) had **not** traveled to an area known to have active transmission of these viruses, immediately alert CDPH and the local vector control agency that the disease may have been locally acquired (which suggests that invasive *Aedes* mosquitoes may be present in the area but not yet detected). The same consideration and response should be given to any other location visited by the patient(s) within the viral infection period (pre- and post-symptom onset).

### Post-Detection of *Aedes aegypti/albopictus*

- Upon initial detection of invasive *Aedes* mosquitoes, collaborate with the local vector control agency in issuing a media release describing the discovery of invasive mosquitoes and the disease risks they present. Encourage residents to use repellent and avoid mosquito bites while at home and while travelling abroad to areas with endemic transmission of dengue, chikungunya, and/or Zika. Residents should continue to take



protective measures against mosquito bites for 3 weeks after their return. Reassure the public that virus transmission risks are low and request that the public contact the local vector control agency regarding any daytime-biting mosquitoes. See media release template, [Appendix B](#).

- Enhance surveillance for human cases of dengue, chikungunya, and Zika by following up as soon as possible with all suspect, probable, and confirmed case-patients for their travel history and by entering all patient information into CalREDIE (for non-participating jurisdictions, report patient information by fax or secure email). Immediately notify CDPH of any patient who had **not** traveled to an area with endemic or current transmission of the virus associated with their infection. Educate case-patients about the importance of wearing repellent and avoiding mosquito bites while they are symptomatic.
- Notify the local vector control agency of any suspect, probable, or confirmed cases of dengue, chikungunya, or Zika infection. Timely notification is critical for enhancing mosquito surveillance and control of adult mosquitoes in the vicinity of the case-patient's residence, particularly within a 100-200 meter radius, to minimize the potential for arbovirus transmission.
  - Advise the local vector control agency of their responsibility to maintain patient confidentiality.
  - The information disclosed to local vector control should be limited to that needed to investigate and control virus transmission by mosquitoes. Minimum information should include travel history (local and international), date of symptom onset, and residential address.
- Educate the local medical community on signs and symptoms of dengue, chikungunya, and Zika infection (see **Appendices C, D, E, and G**) and remind healthcare providers to test and report all potential cases. Dengue, chikungunya, Zika, and yellow fever virus infections are all reportable in California.
  - Provide and disseminate educational materials from CDPH and/or the US Centers for Disease Control and Prevention (CDC). See [Appendix G](#).
  - Provide information on testing suspect patients for infection.
- Assess your local public health laboratory's capacity to test for dengue, chikungunya and Zika viruses. If no capacity exists, specimens can be sent to commercial laboratories or the CDPH Viral and Rickettsial Disease Laboratory (VRDL) for testing. See **Appendices C, D, and E**.
- If notified by the local vector control agency of *Aedes aegypti/albopictus* positive for dengue, chikungunya, or Zika virus:
  - Assume that the finding of infected mosquitoes is indicative of potential local transmission and implement all applicable steps listed in the next section, **Locally Acquired Human Infection(s) Identified**.
  - Immediately enhance surveillance for potential locally acquired human cases of dengue, chikungunya, and Zika infection, starting where positive mosquitoes were collected.
  - Notify the medical community, including hospitals and laboratories, to look for diagnosed and suspected cases of dengue, chikungunya, and Zika infections,

regardless of recent travel history, and to report them as soon as possible. Focus case surveillance in and around areas where infected mosquitoes were collected.

- Consider issuing a media release in coordination with CDPH and the local vector control agency to raise awareness of increased virus transmission potential. If appropriate, state that no locally acquired human cases have been confirmed.

### **Locally Acquired Human Infection(s) Identified**

- Immediately notify the local vector control agency of any suspect, probable, or confirmed locally acquired cases of dengue, chikungunya, or Zika. Provide the information necessary to conduct mosquito surveillance and control to interrupt virus transmission in the target area. Minimum information should include travel history (local and international), date of symptom onset, and residential address.
- Work collaboratively with CDPH and the local vector control agency to issue a joint media release ensuring patient confidentiality. See media release template, [Appendix B](#).
- Conduct epidemiologic investigation and enhanced surveillance where the case-patient(s) spent the most time during the 2 weeks prior to onset of illness (e.g., home, neighborhood, and workplace).
- Work with the local vector control agency to enhance mosquito surveillance and control where exposure to invasive *Aedes* mosquitoes may have occurred, and to distribute public education materials to raise awareness about invasive *Aedes* mosquitoes, the viruses they can transmit, symptoms of disease, and use of personal protective measures.
- Advise patients to take all steps to avoid mosquito bites to minimize the risk of infecting mosquitoes and ongoing local transmission.
- Enhance surveillance for additional locally acquired human cases by notifying the local medical community, including hospitals and laboratories, to look for and encourage testing of all suspected dengue, chikungunya, and Zika infections, regardless of recent travel history, and to report them as soon as possible, potentially through a Health Alert Network (HAN) notification.
- Once local human transmission is documented, follow up promptly on all additional suspect cases of dengue, chikungunya, and Zika infections as potentially locally acquired and notify CDPH via CalREDIE or by secure email or fax.
- Engage the public in detecting and reporting daytime-biting mosquito activity to the local vector control agency, reducing mosquito larval habitats on their property, and protecting themselves from mosquito bites.
- Escalate and expand all activities in parallel with increases in the number of human cases identified.

## Role of the California Department of Public Health

Services available to support local vector control and public health agencies include:

- Development of public education materials on invasive *Aedes* (e.g., distribution maps, list of cities with established populations, brochures, fact sheets, flyers, door hangers) and local media releases
- Development of fact sheets and information for health professionals, including [Information for Clinicians: \*Aedes aegypti\* and \*Aedes albopictus\* Mosquitoes in California and Reporting Patients with Suspected Dengue or Chikungunya to Public Health](#) (see [Appendix G](#))
- Consultation and assistance regarding:
  - Mosquito identification, surveillance techniques, control options, and allocation of available resources
  - Human arbovirus infection symptoms and diagnosis
  - Human arbovirus case testing and evaluation
  - Response to outbreak of human disease
- Interpretation of laboratory-based insecticide resistance test results for *Ae. aegypti* and *Ae. albopictus*
- Facilitation of collaboration and communication among agencies in affected and neighboring counties
- Providing epidemiological information on cases of dengue, chikungunya, Zika, and other mosquito-borne viral infections in California, including via CD Brief, reports, and presentations to stakeholders
- Issuing statewide media releases
- Coordinating and leading the regional or statewide public health response including surveillance, investigation, and control in the event of widespread local arbovirus transmission involving multiple jurisdictions
- Providing back up and/or surge diagnostic laboratory testing of clinical specimens to determine possible dengue, chikungunya, Zika, and other mosquito-borne viral infections and providing technical support for laboratory testing as needed

## Discussion of Recommended Actions for Local Vector Control Agencies

### Mosquito Surveillance

Effective surveillance for *Ae. aegypti* and *Ae. albopictus* requires the careful selection and placement of target-specific traps, larval surveys in unconventional areas, and a much greater level of interaction with the public. The success of these activities depends on understanding the ecology and behavior of these container-breeding mosquitoes to maximize the potential for monitoring and controlling their populations. Field and laboratory staff should be able to identify egg, larval, pupal, and adult stages of these mosquito species and anticipate that they might be collected during routine surveillance operations outside of known infestation areas. Currently available traps may not capture eggs or adults effectively when mosquito populations

are low or patchy in the environment, but the probability of detection can be improved by increasing the number of traps in a given target area. At present, there are no established guidelines for the number of traps (of any type) necessary for a comprehensive *Ae. aegypti* or *Ae. albopictus* surveillance program. See [Appendix A](#) and [Traps and Collection Methods for \*Aedes aegypti\* and \*Aedes albopictus\* Surveillance and Control](#) in [Appendix G](#) for a description and discussion of several of these traps.

## Pre-Detection versus Post-Detection Mosquito Surveillance

Surveillance strategies will vary depending on whether invasive *Aedes* mosquitoes have been detected.

### Pre-Detection of *Aedes aegypti*/*albopictus*

Container-breeding mosquitoes such as *Ae. aegypti* and *Ae. albopictus* are notoriously difficult to control once they become established in residential areas. The best chance for eradicating these mosquitoes from newly infested areas is early detection, before the population becomes abundant and widespread. Local agencies should develop and implement an early detection plan for invasive *Aedes* mosquitoes that employs the use of strategically placed ovi- and adult traps and an outreach program designed to educate and mobilize the public to report daytime-biting mosquitoes. Soliciting public participation is critical because residents are most likely to observe unusual mosquito activity on their own properties, where there may be large numbers of water-holding containers to support larval development.

The mosquito surveillance database maintained by the local vector control agency should be reviewed and, if necessary, modified to accept data on invasive *Aedes* mosquitoes. Data should be maintained locally in a standardized format that allows for easy comparisons of data over time and among geographic locations. All data on invasive *Aedes* collection efforts, including traps or door-to-door surveys that did not find mosquitoes, should be reported in the [California Vectorborne Disease Surveillance Gateway \(CalSurv\)](#) (<https://gateway.vectorsurv.org/>). To avoid redundant entry, agencies with in-house data systems may exchange data automatically with the VectorSurv Gateway using web services. For questions or assistance, contact the VectorSurv Gateway development team ([help@vectorsurv.org](mailto:help@vectorsurv.org)).

The potential routes of invasive *Aedes* mosquito introduction into an area need to be considered and a portion of the early detection efforts focused on these entry routes. Historically, commercial importation of certain goods (e.g., live plants, used tires) facilitated introductions; however, with the current widespread infestations of invasive *Aedes* in California, residents and visitors are now more likely to unintentionally transport and introduce these mosquitoes to new locations. The type and number of sites selected for surveillance will be determined by the local agency's resources but should include both commercial and residential areas where locations such as cemeteries, plant nurseries, and neighborhoods may have an abundance of water-holding containers.

- **Traps.** Placement of traps should be carefully considered to maximize the likelihood of detection. There is currently no established formula for determining the best traps to

use, the ideal number of traps, or trap placement for any given area. However, the known advantages, disadvantages, and performance of different trap types (**Appendices A** and **G**) suggest that using more than one trap type and using as many as economically feasible should increase the chance of detecting invasive mosquitoes. Trap inspections and maintenance can be extended to approximately one-week intervals to optimize the use of resources.

- **Public Education and Outreach.** Educating the public about invasive mosquitoes and instructing people to report any suspicious sightings or daytime biting annoyance is crucial for early detection. The outreach program should include educational materials that are culturally and linguistically appropriate to fit the diversity of the local community and target residential, commercial, and industrial sectors. The program can include written and electronic materials available at the agency's headquarters and website, flyers for distribution to homes and businesses, roadway billboards, ads on public transportation vehicles, workshops, social media, and oral presentations. Information can also be provided to the media to prompt news coverage. Public education and outreach activities have the dual benefit of increasing the chances of early detection while also increasing awareness of local vector control services.

### **Initial Detection of *Aedes aegypti*/*albopictus***

The discovery of *Ae. aegypti* or *Ae. albopictus* in a new area of California should be reported immediately to CDPH. If needed, specimens or photos can be sent to CDPH to confirm identification, and any observations and findings, potential infestation areas, and possible introduction pathways should be shared and discussed. Upon species confirmation, the local vector control agency should initiate their response plan beginning with the notification of the local health department, neighboring vector control agencies, and other agencies as appropriate. Public education materials regarding the discovery should be released at this time, either independently or as joint efforts with the health department or other local agencies, according to previously established plans. Materials should include a media release urging the public to eliminate sources of standing water on their property and report any daytime-biting mosquitoes to the local vector control agency.

Surveillance activities following the discovery of invasive *Aedes* mosquitoes should be intensive and rigorous to provide a comprehensive assessment of population size, geographical spread, and control effectiveness. Rapid surveillance of larger areas can be accomplished by focusing first on presence rather than abundance of invasive mosquitoes, i.e., no need to identify more than one specimen per property. If notified by the local health department of travel-associated *Aedes*-borne infections within the surveillance area, enhanced efforts should be focused near the patient's residence to determine if a risk of local transmission exists.

- **Traps.** The number and variety of traps should be increased relative to pre-detection levels and placed in the areas surrounding the site(s) of discovery to assess the abundance and distribution of invading mosquitoes. Additional traps should be placed outward from identified infestation areas to determine the geographical extent of the population. It should **not** be assumed that the index location (first site where invasive

mosquitoes were discovered) is the initial site of introduction. To aid in these assessments, inspection intervals should be increased to every 1-3 days.

- **Public Education and Outreach.** All aspects of the education and outreach program should be intensified throughout the jurisdictional area of the agency, but particularly in the urban areas surrounding the point(s) of discovery and other known infested areas. Door-to-door campaigns should be initiated immediately to inform and educate residents about the invasive *Aedes* mosquitoes, how they can minimize habitat on their property, and encourage people to report daytime-biting mosquitoes. The door-to-door campaign will also provide an opportunity for additional larval and adult (e.g., observing host-seeking adult females) surveillance on the property, providing initial indications of mosquito abundance and spread.

## Mosquito Control Response

The successful control of invasive *Aedes* mosquitoes depends on a number of factors, especially if eradication from a local area is an objective. Consider the following:

- Proactive planning and preparation are critical prior to the discovery of *Ae. aegypti* or *Ae. albopictus* to ensure a rapid and smooth transition from routine vector control activities to the targeted surveillance and control of invasive *Aedes*.
- Mutual aid agreements previously made with neighboring vector control agencies can be of great assistance in conducting certain aspects of the mosquito surveillance and control response, especially with door-to-door campaigns and ground-based insecticide applications.
- Area warrants for property inspections and control operations are sometimes necessary, therefore the process should be in place for rapid implementation.
- Public education and outreach programs and door-to-door surveillance activities not only provide important information on abundance and spread of invasive *Aedes* mosquitoes, but also aid control in urban environments by reducing potential larval habitats.
- A combination of physical, biological, and chemical control approaches should be used against immature and adult invasive *Aedes* mosquitoes. For thorough implementation, these control activities frequently require the collaboration and cooperation of residential property owners established during education, outreach, and door-to-door campaigns.
- In addition to containers (e.g., jars, pots, bird baths, rain barrels), invasive *Aedes* mosquitoes will often utilize other larval habitats such as relatively small subsurface habitats (e.g., catch basins, dry wells, yard drains, storm water treatment devices, public utility vaults), surface pools (e.g., neglected ponds, water-holding surface depressions in lawns), and vegetation (e.g., tree holes, bromeliad leaf axils).
- The mosquito control response should always be guided by both mosquito and human disease data generated by local surveillance programs.

Surveillance data is crucial for directing control efforts and should be used to continually focus and re-focus resources and control efforts. Critical data includes the presence, relative

abundance, and distribution of invasive *Aedes* mosquitoes as well as information from the local health department regarding suspect, probable, and confirmed cases of dengue, chikungunya, or Zika infections. The primary emphasis of the mosquito control program should be to protect the public from potential virus transmission. This is enhanced by educating and mobilizing the public to implement physical controls to eliminate immature mosquito development on their property and protecting themselves from mosquito bites. When attempting to eradicate a newly detected infestation or when responding to travel-associated or locally acquired human arbovirus infections, it is crucial that local agency staff gain access for inspection of every property in an affected area, including vacant properties and properties with uncooperative owners/residents. A single neglected property can provide the habitat necessary for intensive mosquito production, thus allowing rapid re-invasion or counteracting ongoing control efforts. That same property can also serve as a refuge for potentially infected female mosquitoes to drive local arbovirus transmission. Local vector control agencies may obtain an inspection and abatement warrant, per California Health and Safety Code Section 2053, and or coordinate with law enforcement or other local code enforcement agency to access properties.

EPA-registered biological and chemical control products labeled for larval and adult mosquitoes in California can be used effectively against invasive *Aedes* mosquitoes, but some may require the use of equipment and application techniques not normally employed for the control of indigenous species. Insecticides should be applied only when surveillance data confirm the presence of invasive *Aedes* and indicate a need for control based on factors including mosquito abundance, biting pressure, and risk of disease transmission. Treatment efficacy should be monitored via pre- and post-trapping surveys. Cage trials, when feasible, can provide additional data on product and application efficacy and potential for insecticide resistance. Pesticide resistance profiles of local *Ae. aegypti* or *Ae. albopictus* populations should be monitored periodically to inform the selection of effective active ingredients or potential for diminishing efficacy over time. Treatment options are outlined below; none should be expected to provide long-term control of invasive *Aedes* mosquitoes without removal of larval habitats. In addition, the structural complexity of many urban environments may preclude effective insecticide penetration into the target area even when meteorological conditions appear ideal.

- Formulations of larvicidal products containing active ingredients such as methoprene and *Bacillus thuringiensis* var. *israelensis* can be broadcast into urban environments using ground-based or aerial spray equipment calibrated to produce larger droplet sizes than typical adulticide applications.
- Residual adulticide sprays can be applied using hand-held equipment over small areas (individual properties) to vegetation and other surfaces where adult mosquitoes might take refuge or rest.
- Ultra-low volume (ULV) adulticides can be used to control adult mosquitoes using hand-held or backpack equipment, or over larger areas using truck-mounted foggers when environmental conditions are appropriate.
- Aerial adulticide applications may be considered over urban areas too large to treat efficiently and effectively using ground-based equipment, especially under conditions of a human disease outbreak when adult mosquito numbers require rapid knockdown to interrupt the virus transmission cycle.



Several alternatives to insecticide-based control of invasive *Aedes* are emerging and undergoing field trials in California and elsewhere. These include Wolbachia-based methods, genetically modified mosquitoes, and sterile male releases. These technologies may lead to future proactive options for controlling mosquito populations, but do not currently provide the rapid knock-down of adult female mosquitoes necessary to interrupt virus transmission.

## **Detection of Dengue, Chikungunya, or Zika virus in *Aedes* Mosquitoes**

An important component of many mosquito surveillance programs is testing field-collected females for arbovirus infection. In the case of invasive *Aedes* mosquitoes, females rarely test positive for dengue, chikungunya, or Zika viruses. No dengue-positive mosquitoes were detected during the extensive mosquito surveillance efforts following the detection of locally acquired dengue cases in 2023 and 2024. Therefore, it is left to the discretion of the local vector control agency on whether resources allow the addition of this surveillance element. When successful, however, positive findings may indicate that local transmission has occurred or may occur, that human infections exist even if not reported, and that the virus may be circulating among mosquitoes and humans at a low level. Positive mosquitoes should trigger an immediate surveillance and control response to reduce risk of transmission to local residents and visitors as described in the subsequent section. The local health department also should be notified to enhance human case surveillance. Consider issuing a media release in coordination with CDPH and the local health department to raise awareness of increased virus transmission potential. If appropriate, state that no locally acquired human cases have been confirmed.

## **Travel-associated and Locally Acquired Human Infection(s) Identified**

Human cases of dengue, chikungunya, and Zika are investigated by local health departments and CDPH. A patient's travel history is established to determine whether a person likely acquired the infection from recent travel to an area with ongoing disease transmission or locally. Confirming and classifying human cases can be a lengthy process, and often local health departments receive information weeks after date of symptom onset. During this time, there is the potential that local human-mosquito-human transmission cycles have already begun in the vicinity of the case patient residence or in other places the person may have visited while viremic. Therefore, the local vector control response should be initiated immediately upon notification of any laboratory positive or presumptive-positive cases, regardless of whether classified as travel-associated or locally acquired. The mosquito surveillance and control response should be intensified in areas where potentially viremic persons may have been bitten by *Ae. aegypti* or *Ae. albopictus* mosquitoes to minimize the potential for local disease transmission. If additional locally acquired human cases are identified in the area, a more aggressive response (e.g., increasing the size of the target area in response to local data) should be implemented in consultation with CDPH and in coordination with the local health department and other appropriate agencies. The response will escalate further if widespread local transmission is detected and regional coordination may become necessary. Local or regional outbreaks may require a more intensive mosquito control response including aerial spraying. When multiple jurisdictions are involved, CDPH may coordinate and lead the regional public health response including surveillance, investigation, and control.



## Discussion of Recommended Actions for Local Health Departments

### Human Disease Surveillance

To date, none of the exotic arboviruses carried and transmitted by *Ae. aegypti* and *Ae. albopictus* are known to be continuously circulating among mosquitoes in California and the risk of these viruses being introduced into established populations of invasive *Aedes* from infected visitors and returning travelers is currently considered to be low; however, a single viremic person with dengue, chikungunya, or Zika who is subsequently bitten by a female *Ae. aegypti* or *Ae. albopictus* could start local disease transmission within a community. There are several conditions and a sequence of events that would need to be in place for local transmission of dengue, chikungunya, or Zika to occur.

These include:

1. An infected and viremic individual would need to return to a locality in California where there are *Ae. aegypti* and/or *Ae. albopictus* mosquitoes. The viremic period is typically 7 days, beginning 1-2 days before symptom onset. Up to 80% of individuals infected with dengue or Zika will remain asymptomatic during their viremic period, along with approximately 2/3 chikungunya infections. If the infected person returned more than a week after onset of illness, then transmission of virus from this person is less likely.
2. A female mosquito would need to bite the infected person while this person is viremic.
3. The mosquito would need to live approximately 10-11 days after taking a virus-infected blood meal to allow for the virus to multiply and migrate to the salivary glands (extrinsic incubation period); most adult mosquitoes live < 14 days, making subsequent virus transmission unlikely, but this is dependent on many environmental and ecological factors.
4. The infected mosquito would need to bite one or more susceptible persons who become infected and then viremic, but who may or may not become symptomatic. Both *Ae. aegypti* and *Ae. albopictus* typically take multiple blood meals during each gonotrophic cycle (blood ingestion and egg development cycle; 2-7 day intervals) and therefore an infectious female may bite multiple people over a short period of time.
5. This cycle would need to be repeated for sustained transmission to occur.

Note that detection of locally acquired human infection with dengue, chikungunya, or Zika virus may occur prior to the discovery of invasive *Aedes* mosquitoes in an area.

### Pre-Detection of *Aedes aegypti/albopictus*

Detection and reporting of suspect, probable, or confirmed human infections with dengue, chikungunya, or Zika viruses is critical for monitoring the possible points of introduction of these pathogens into California and controlling the spread of disease in the event of an outbreak. All infections, regardless of status (i.e., suspect, probable, or confirmed), should be reported using the real-time, secure web-based CalREDIE system maintained by CDPH. Non-participating jurisdictions should report all infections by submitting the appropriate paper case report form by secure email or fax immediately after the investigation is complete. Dengue,

chikungunya, Zika, and yellow fever virus infections are all reportable in California. The surveillance case definitions and laboratory testing for dengue (i.e., dengue and severe dengue), chikungunya, and Zika are summarized in **Appendices C, D, and E**, respectively. **Appendix G** contains resources for more information on dengue, chikungunya, Zika, and invasive *Aedes* mosquitoes.

Reports associated with human arboviral infections should include information regarding symptom onset date and travel history to elucidate if infections were acquired outside of California or locally. If the case-patient had no travel history to areas endemic for the disease within the incubation period, and for Zika cases no sexual contact with a returned traveler, CDPH and the local vector control agency should be contacted immediately. The local health department should ensure that patient confidentiality is maintained regarding sharing of personal identifiers (e.g., name, address, laboratory test results). The absence of travel suggests that the infection may have been acquired locally even if the person resides in an area not known to be infested with *Ae. aegypti* or *Ae. albopictus*. Invasive mosquitoes can be elusive in the environment and can be associated with relatively small habitats (e.g., residential backyards). The local vector control agency should conduct a follow-up investigation of the general area surrounding the case-patient's residence to determine if invasive *Aedes* mosquitoes are present, but previously undetected.

A public education plan should be prepared and include a media release to be implemented if invasive *Aedes* mosquitoes are detected within the jurisdiction of the local health department. A similar response plan should be prepared in the event that local transmission of dengue, chikungunya, or Zika virus is confirmed. Both plans should include: 1) a local health advisory to the medical community to increase awareness of exotic mosquito-borne viral infections in humans (the advisory should specify whether a locally acquired human case has been detected and recommendations should be tailored accordingly) and 2) a request for the public to report daytime-biting mosquitoes, minimize habitats suitable for invasive *Aedes* mosquitoes, protect themselves from mosquito bites, and recognize common symptoms of dengue, chikungunya, and Zika disease. Coordination with the local vector control agency, or CDPH where no local vector control agency exists, ensures that messages and materials distributed to the public and to the media remain consistent. Response plans can be administered independently or jointly with the local vector control agency.

### **Post-Detection of *Aedes aegypti*/*albopictus***

Once *Ae. aegypti* and *Ae. albopictus* mosquitoes are established in an area, visitors and returned travelers infected with dengue, chikungunya, or Zika virus may infect local *Aedes* mosquitoes if they are bitten while viremic. The previously established public education plan regarding the discovery of invasive *Aedes* mosquitoes should be initiated. Local health departments should ensure that the local medical community is educated about the exotic arboviral disease risks associated with *Ae. aegypti* and *Ae. albopictus*, signs and symptoms of these diseases in humans, human specimen collection for laboratory confirmation and clinical diagnosis, proper patient treatment, and disease reporting.

The local vector control agency should be notified promptly of any suspect, probable, or confirmed cases of dengue, chikungunya, or Zika within their jurisdictional area. Patient confidentiality should be maintained. The prevention or reduction of transmission of *Aedes*-borne viruses is completely dependent on the control of mosquitoes and limiting person-mosquito contact.

### **Detection of Dengue, Chikungunya, or Zika Virus in Invasive *Aedes* Mosquitoes**

Locally collected invasive *Aedes* mosquitoes that test positive for dengue, chikungunya, or Zika virus are an indicator of local transmission in the area surrounding a trap location. Mosquitoes infected with virus suggest that local transmission has occurred or may occur, that human infections exist even if not identified, and that the virus may be circulating among mosquitoes and humans at a low level. A positive mosquito finding should trigger an immediate mosquito surveillance and control response by the local vector control agency to reduce risk of transmission to local residents and visitors.

If notified of a positive mosquito sample, human case surveillance should be enhanced in the vicinity of the mosquito collection. The local health department should coordinate with the local vector control agency and CDPH to follow-up on any human arbovirus cases subsequently identified as residing in the vicinity of the positive sample. The medical community should be notified of the increased risk of exotic arboviral infection and noting if locally acquired infection(s) have been identified in the area. The public should be advised to use mosquito bite prevention measures, and the medical community encouraged to consider conditions in patients with compatible illness or travel history and report promptly all suspect, probable, or confirmed cases of dengue, chikungunya, or Zika infection. Consider issuing a media release in coordination with CDPH and the local vector control agency to raise public awareness of increased virus transmission potential. If appropriate, state that no locally acquired human cases have been confirmed.

### **Locally-Acquired Human Infection(s) Identified**

The discovery of one or more human infections of dengue, chikungunya, or Zika virus that potentially was locally acquired should be addressed aggressively and immediately. To identify possible additional cases in an area where a locally acquired case may have been exposed to infected mosquitoes, an epidemiologic investigation and enhanced surveillance should be implemented to cover the area(s) where the case-patient spent the most time (e.g., home, neighborhood, and workplace) within the 2 weeks leading to onset of illness. Local vector control agencies and CDPH should be notified to ensure that mosquito surveillance and control of adult mosquitoes is enhanced around the residence and any areas the identified case-patient may have been exposed to biting mosquitoes during their viremic period. Patients should be advised to take all steps to prevent mosquito bites to reduce the risk of spread to local mosquito populations. Zika case-patients should be advised to take measures to avoid sexual transmission to partners. The previously developed public education plan should be initiated. Additional response efforts could include:

- Facilitated testing of suspect cases and enhanced case finding

- Additional coordination between local and state public health epidemiologists and public health laboratorians
- Enhanced coordination and communication with clinical diagnostic laboratories
- Outreach and education to healthcare providers on the diagnosis and clinical management of dengue, chikungunya, and Zika viruses
- An enhanced media campaign to the public

All activities should escalate in the event of widespread local transmission, and, if multiple jurisdictions are involved, CDPH may coordinate and lead the regional public health response including surveillance, investigation, and control. Neighboring jurisdictions, states, and the CDC should also be notified, depending on the extent of disease transmission.

The implications of local transmission of exotic mosquito-borne viruses are many and require the greatest level of response. Close and rapid interagency communication with CDPH and the local vector control agency is critical to ensure rapid suppression of adult invasive *Aedes* mosquitoes to break the human-mosquito-human disease cycle and prevent outbreaks of dengue, chikungunya, or Zika.

## Appendix A

### Examples of Target-Specific Traps for Invasive Container-Breeding Mosquitoes such as *Aedes aegypti* and *Aedes albopictus*

#### Ovitrap

Advantages	Disadvantages
<ul style="list-style-type: none"><li>• Inexpensive</li><li>• Easy to deploy, inspect, and refresh</li><li>• Inspection intervals can be up to 7 days</li><li>• Eggs can be reared to adults for use in pesticide resistance assays</li></ul>	<ul style="list-style-type: none"><li>• Requires that eggs be reared in the laboratory to confirm species identification</li><li>• Ovitrap can support mosquito production if left in the environment for more than 7 days or lost</li><li>• Success may be influenced by availability of competing container habitats</li><li>• May be tipped over, dry out, or flooded by rainfall or other environmental factors</li></ul>

The ovitrap is the most basic surveillance tool for *Ae. aegypti* and *Ae. albopictus* in the urban environment. In general, an ovitrap consists of a small dark-colored container (e.g., 24-32 oz black plastic cup) partially filled with water or mild attractant infusion and with an oviposition medium (e.g., wood tongue depressor, germination paper, construction paper). Female mosquitoes seeking an egg-laying site may choose to deposit some eggs on the oviposition medium provided in the cup. Almost any small container can be used as an ovitrap, but studies have found black-colored containers to have superior performance.

#### Limitations

- Detection success may be directly dependent on the number of ovitraps deployed. For example, a city block with one ovitrap per property may increase the likelihood of detecting presence of *Ae. aegypti* and *Ae. albopictus* than the same city block with only one deployed ovitrap.
- Does not provide quantitative information on the abundance of adults in the environment; only evidence of the presence of at least one adult female.

## CDC-AGO (Autocidal Gravid Ovitrap)

Advantages	Disadvantages
<ul style="list-style-type: none"><li>• Inexpensive</li><li>• Easy to deploy, inspect, and refresh</li><li>• Inspection intervals can be lengthened; the trap may function for more than 8 weeks without need for maintenance</li><li>• The design prevents adult access to standing water, thus rarely will support mosquito production if left unattended</li><li>• Removes egg-laying females from the environment</li><li>• Allows immediate identification of captured adults</li><li>• Can provide some information on the relative abundance of adults in a given environment.</li></ul>	<ul style="list-style-type: none"><li>• Larger, bulkier, and heavier than standard ovitraps</li><li>• More visible in the environment</li><li>• Adults trapped by the adhesive may be difficult to dislodge for identification and may not be suitable for testing for viruses or pesticide resistance</li><li>• Glue paper maintenance frequency may vary depending on air-borne debris, non-target insect captures, and relative humidity of trap site</li></ul>

Several variants of “lethal ovitraps” similar in concept to the CDC-AGO have been developed. The concept behind these traps is to lure oviposition-site-seeking females to a container from which they cannot escape or where they come into contact with a lethal dose of insecticide. The AGO Trap is made from modified 1 gallon and 5 gallon black plastic utility buckets partially filled with a hay-based infusion. Female mosquitoes seeking an egg-laying site can enter part-way into the bucket through an opening but are blocked from accessing the water by a screen. An adhesive on the vertical surface of the entrance captures mosquitoes on contact.

### Limitations

- Detection success may be directly dependent on the number of AGO traps deployed in a given area.

## Biogents (BG) Sentinel Adult Trap

Advantages	Disadvantages
<ul style="list-style-type: none"> <li>• Target-specific trap developed for capture of adult <i>Ae. aegypti</i> and <i>Ae. albopictus</i>, with option to add an attractant lure (human scent lure, octenol, or CO<sub>2</sub>).</li> <li>• Does not require a lure to attract <i>Ae. aegypti</i> and <i>Ae. albopictus</i></li> <li>• Few non-target species are attracted to these traps unless CO<sub>2</sub> attractant is added to the system</li> <li>• Can be plugged into available 110V outlets for continuous operation if desired to increase inspection intervals</li> <li>• Captures both males and females</li> <li>• Allows immediate identification of captured adults</li> <li>• Can provide some information on the relative abundance of adults in a given environment</li> <li>• Trapped live mosquitoes can be tested for arbovirus if traps are serviced frequently (e.g., every 1-2 nights)</li> </ul>	<ul style="list-style-type: none"> <li>• Expensive</li> <li>• Selection of suitable deployment areas safe from theft, vandalism, and weather/environmental damage can be time-consuming</li> <li>• Battery packs discharge rapidly, usually in less than 3 days</li> <li>• Trapped mosquitoes can escape from the net bag of older models if the power supply is disconnected or discharged, or if the fan motor fails. Newer models provide a trapdoor to minimize escape in the event of a fan failure</li> <li>• Ants may damage or remove mosquitoes from trap</li> </ul>

The BG Sentinel is an adult trap that selectively targets *Ae. aegypti* and *Ae. albopictus* and is currently considered one of the most effective commercially available adult traps for these two species. Both males and females may be attracted to the trap and are captured by a suction fan into a small net bag. The design is versatile in that commercially available lures or CO<sub>2</sub> can be incorporated into the body of the trap to improve attractiveness.

### Limitations

- Detection success may be directly dependent on the number of BG Traps deployed in a given area.

## Appendix B

### Media Release Templates

*Example Vector Control Agency (VCA) / Local Health Department joint press release subsequent to first detection of an invasive Aedes mosquito*

*Aedes aegypti* Mosquito found in [City, County]  
(Substitute *Aedes albopictus* for *Aedes aegypti* as appropriate.)

[City] – The [VCA] has detected *Aedes aegypti* mosquitoes at/in [area]. The first detection was on [date]. [VCA] is working with the [City, County] Department of Health to evaluate the extent of the infestation and will aggressively target problem areas to prevent its spread.

*Aedes aegypti* is a small (about ¼ inch) black and white mosquito that bites aggressively during the day. *Aedes aegypti* is not native to California; however, it is a common mosquito in some urban areas of the southeastern United States and Arizona and has become established in many [California counties](#). *Aedes aegypti* has the potential to transmit several viruses including dengue, chikungunya, and Zika. These viruses are not currently found in California.

"Our goal is to control and eliminate this mosquito population," said [VCA Manager]. "We are doing everything to help ensure this mosquito does not become established in our communities."

The [VCA] has expanded surveillance efforts for this type of mosquito. [Text example: The District has deployed a variety of traps for adult mosquitoes and mosquito eggs surrounding the location where *Aedes aegypti* was found. Additionally, District staff are conducting door-to-door inspections of properties for mosquito breeding and standing water at homes near *Aedes aegypti* detections].

[Insert if relevant - This mosquito was previously found in [area or county] in [year] near [place] but was successfully eradicated by the [VCA] and did not become established here].

The public can play a critical role in helping to control the spread of this mosquito. *Aedes aegypti* lays its eggs just above the water line in small containers and vessels that hold water, such as dishes under potted plants, bird baths and feeders, ornamental fountains, tin cans, children's toys, discarded tires, and yard drains. It's important for residents to look around their yard and outside their home and dump out even the smallest amount of standing water. Clean and scrub bird baths and pet watering dishes weekly and dump the water from overflow dishes under potted plants.

[County] Health Officer [Name] reminds people to do the following to reduce the chances of being bitten by mosquitoes:

- Apply repellents containing EPA-registered ingredients such as DEET, picaridin, oil of lemon eucalyptus, or IR3535 to exposed skin and/or clothing (as directed on the product label).



- Wear long sleeve shirts, long pants, socks, and shoes when mosquitoes are most active.
- Be sure window and door screens are in good repair to prevent mosquitoes from entering your home.

Residents experiencing mosquito bites during the day should report them immediately to [VCA contact info]

If you are sick with fever, headache, and joint or muscle pain after returning from an area where dengue, chikungunya, or Zika occurs, contact your doctor or health care provider, and prevent mosquito bites by using mosquito repellent or staying indoors as much as possible while sick to help prevent possible spread of the virus to mosquitoes in the area.

Additional information on *Aedes* [species] can be found at:

[Local health department website]

[VCA website]

[California Department of Public Health \*Aedes\* Mosquitoes webpage](#)

*Example Local Health Department [LHD] press release subsequent to first detection of a locally acquired human case of dengue. If this template is used for another locally acquired exotic mosquito-borne disease, such as chikungunya or Zika, please edit the paragraph describing symptoms.*

--First Confirmed Locally Acquired Dengue Case in [County]

[City/County] – Today, the [County] Health Department announced that the first locally acquired human dengue case has been confirmed in a [county] resident. [if applicable: To date, (number) locally acquired dengue cases have been previously detected in California].

Dengue (pronounced den' gee) is caused by a virus that is transmitted to humans by the bite of an infected *Aedes aegypti* or *Aedes albopictus* mosquito. *Aedes* mosquitoes have been found in [cities] in [county]. Dengue virus cannot be transmitted from person to person. Symptoms of dengue may include high fever, severe headache, pain behind the eyes, joint pain, and rash. Health care providers should contact the [County] Health Department if they suspect an individual may have dengue or another mosquito-borne illness.

The [Vector Control Agency - VCA] and the [LHD] are enhancing surveillance, prevention, and mosquito control efforts. Residents should take basic precautions to protect themselves from mosquitoes by following the Department of Health recommendations. [County] Health Officer [Name] reminds people to do the following to reduce their chances of being bitten by mosquitoes and to help prevent spread of the virus:

- Apply repellents containing EPA-registered ingredients such as DEET, picaridin, oil of lemon eucalyptus, or IR3535 to exposed skin and/or clothing (as directed on the product label).
- Wear long sleeve shirts, long pants, socks, and shoes when mosquitoes are most active.
- Be sure window and door screens are in good repair to prevent mosquitoes from entering your home.
- Residents experiencing mosquito bites during the day should report them to [VCA contact info] and should contact their health care provider if they have dengue-like symptoms.

If you are sick with fever, headache, and joint or muscle pain, contact your doctor or health care provider and prevent mosquito bites by using mosquito repellent or staying indoors as much as possible while sick to help prevent possible spread of the virus to mosquitoes in the area.

Additional information on dengue and *Aedes* [species] can be found at:

[Local health department website]

[VCA website]

[California Department of Public Health \*Aedes\* Mosquitoes webpage](#)

## Appendix C

### Dengue Surveillance Case Definition, Reporting, and Laboratory Testing

#### Clinical Description (Dengue, Severe Dengue)

*Dengue:* Dengue is most commonly an acute febrile illness defined by the presence of fever and one or more of the following: nausea/vomiting, rash, aches and pains (headache, retro-orbital or ocular pain, joint pain, muscle pain), leukopenia, positive tourniquet test, or any warning signs of severe dengue (persistent vomiting, extravascular fluid accumulation (e.g., pleural or pericardial effusion, ascites), mucosal bleeding at any site, liver enlargement >2 centimeters, or increasing hematocrit concurrent with rapid decrease in platelet count).

*Severe Dengue* is characterized by any of the following:

- Severe plasma leakage evidenced by hypovolemic shock and/or extravascular fluid accumulation (e.g., pleural or pericardial effusion, ascites) with respiratory distress. A high hematocrit value for patient age and sex offers further evidence of plasma leakage.
- Severe bleeding from the gastrointestinal tract (e.g., hematemesis, melena) or vagina (menorrhagia) that requires medical intervention such as intravenous fluid resuscitation or blood transfusion.
- Severe organ involvement, including any of the following: elevated liver transaminases (aspartate aminotransferase (AST) or alanine aminotransferase (ALT)  $\geq 1,000$  per liter (U/L)), impaired level of consciousness and/or diagnosis of encephalitis, encephalopathy, or meningitis, or heart or other organ involvement including myocarditis, cholecystitis, and pancreatitis.

#### Laboratory Criteria for Classification

*Confirmatory:* Any one of the following:

- Isolation of dengue virus from or demonstration of specific arboviral antigen or genomic sequences in serum, plasma, blood, cerebrospinal fluid (CSF), or other body fluid or tissue by cell culture, reverse-transcriptase polymerase chain reaction (RT-PCR) test, immunofluorescence or immunohistochemistry.
- Detection in serum or plasma of DENV NS1 antigen by a validated immunoassay.
- Seroconversion from negative for dengue virus-specific serum immunoglobulin M (IgM) antibody in an acute phase ( $\leq 5$  days after symptom onset) specimen to positive for dengue-specific serum IgM antibodies in a convalescent-phase specimen collected  $\geq 5$  days after symptom onset.
- Seroconversion or demonstration of a  $\geq 4$ -fold rise in reciprocal immunoglobulin G (IgG) antibody titer to dengue virus antigens in serum samples collected  $>2$  weeks apart, **and** confirmed by a neutralization test (e.g., plaque reduction neutralization test) with a  $>4$ -fold higher end point titer as compared to other flaviviruses tested.

*Presumptive/Probable:*

- A positive dengue-specific IgM antibody test, on a single acute or convalescent phase serum specimen.

*Suspect:*

- The absence of IgM anti-DENV by validated immunoassay in a serum or CSF specimen collected < 5 days after illness onset and in which molecular diagnostic testing was not performed in a patient with an epidemiologic linkage.

**Epidemiologic Linkage**

- Travel to a dengue endemic country or presence at location with ongoing outbreak within previous two weeks of dengue-like illness, **or**
- Association in time and place with a confirmed or probable dengue case.

**Case Classification**

*Suspected:* A clinically compatible case of dengue, or severe dengue with an epidemiologic linkage.

*Probable:* A clinically compatible case of dengue, or severe dengue with laboratory results indicative of probable infection.

*Confirmed:* A clinically compatible case of dengue, or severe dengue with confirmatory laboratory results.

**Dengue Reporting**

All infections, regardless of status (i.e., suspect, probable, or confirmed) should be reported using the real-time, secure web-based California Reportable Disease Information Exchange (CalREDIE) system maintained by CDPH. Non-participating jurisdictions should report all dengue by submitting the paper dengue case report form by secure email or fax immediately after the investigation is complete. For cases in which no travel history is indicated or local transmission is suspected, CDPH should be notified immediately by telephone.

**Dengue Laboratory Testing**

Dengue viruses are members of the family Flaviviridae and have sufficient antigenic similarity to Zika virus, yellow fever virus, Japanese encephalitis virus, and West Nile virus that previous infection with or vaccination against another flavivirus may raise cross-reactive serum antibodies. After a primary infection with a heterologous flavivirus, subsequent antibody testing by ELISA may produce false positive results for a different flavivirus. The plaque reduction neutralization test (PRNT) may resolve cross-reactive serum antibodies and identify the infecting virus; however, in individuals with multiple previous flavivirus infections, PRNT may not differentiate among different flaviviruses. This demonstrates the complexity inherent in serological diagnosis and differentiation in populations living in regions where more than one flavivirus co-circulates. However, only a small proportion of the U.S. population has evidence of

previous flavivirus infection (or vaccination) so that cross-reactive flavivirus antibodies should not be a significant limitation to dengue diagnosis among most U.S. travelers. Among U.S. residents, most testing for dengue is done through private clinical laboratories using IgM or IgG detection techniques.

Serologic (IgG and IgM) and molecular assays to detect evidence of dengue virus infection are available through commercial laboratories, many local public health laboratories, as well as from the CDPH Viral and Rickettsial Disease Laboratory (VRDL). Testing may include:

- *EIA or IFA for IgM and IgG antibodies.* Serologic assays do not distinguish among dengue serotypes and may be cross-reactive with other flaviviruses. When a positive detection is made for dengue, VRDL can perform a plaque reduction neutralization assay (PRNT) to attempt to distinguish between dengue and other exotic (e.g., Zika virus) or endemic flaviviruses (i.e., West Nile virus, St. Louis encephalitis virus).
- *Real-time RT-PCR (RT-qPCR) for acute serum, plasma, or blood specimens.* Some RT-qPCR tests will discriminate among the four dengue serotypes. Blood for RT-qPCR should be collected within 14 days of symptom onset.
- *NS1 antigen testing.* These tests detect the non-structural protein 1 (NS1) of dengue virus in serum samples collected during the first 7 days of illness. They can confirm dengue virus infection, but cannot provide serotype information.

Acute samples that test positive for dengue-virus specific antibodies at commercial laboratories should prompt ordering of convalescent testing. Both acute and convalescent samples should be forwarded to VRDL for confirmatory testing.

Samples may be submitted to VRDL using the *General Purpose Specimen Submittal Form* available on the [VRDL Specimen Shipping Guidelines and Current Specimen Submittal Forms webpage](#). Details on orderable tests and specimen requirements are available in the [VRDL Laboratory Test Catalog](#).

#### **VRDL Contact Information**

Main Telephone Number	(510) 307-8585
Fax Number	(510) 307-8599

Shipping Address (for hand delivery or private carriers):

Viral and Rickettsial Disease Laboratory  
Attn: Specimen Receiving  
850 Marina Bay Parkway  
Richmond, CA 94804

Reference testing is available from CDC's Dengue Branch, Division of Vector-Borne Infectious Diseases, National Center for Infectious Diseases:

1324 Cañada St.,  
Puerto Nuevo,

San Juan, PR 00920-3860,  
Telephone 787-706-2399  
Fax 787-706-2496

To arrange for reference testing at CDC, contact your local public health department.

## Appendix D

### Chikungunya Surveillance Case Definition, Reporting, and Laboratory Testing

#### Clinical Description (Chikungunya Fever)

*Chikungunya Fever:* Chikungunya fever is characterized by **all** of the following:

- Fever or chills as reported by the patient or a health-care provider.
- Absence of a more likely clinical explanation.

Chikungunya is most often characterized by acute onset of fever (typically  $>39^{\circ}\text{C}$  [ $102^{\circ}\text{F}$ ]) and polyarthralgia. Joint symptoms are usually bilateral and symmetric, and can be severe and debilitating. Other symptoms may include headache, myalgia, arthritis, conjunctivitis, nausea/vomiting, or maculopapular rash. Clinical laboratory findings can include lymphopenia, thrombocytopenia, elevated creatinine, and elevated hepatic transaminases.

Acute symptoms typically resolve within 7-10 days. Rare complications include uveitis, retinitis, myocarditis, hepatitis, nephritis, bullous skin lesions, hemorrhage, meningoencephalitis, myelitis, Guillain-Barré syndrome, and cranial nerve palsies. Persons at risk for severe disease include neonates exposed intrapartum, older adults (e.g.,  $>65$  years), and persons with underlying medical conditions (e.g., hypertension, diabetes, or cardiovascular disease). Some patients might have relapse of rheumatologic symptoms (e.g., polyarthralgia, polyarthritis, tenosynovitis) in the months following acute illness. Studies report variable proportions of patients with persistent joint pains for months to years. Mortality is rare and occurs mostly in older adults. The majority of people infected with chikungunya virus become symptomatic. The incubation period is typically 3-7 days (range, 1-12 days).

#### Laboratory Criteria for Classification

*Confirmatory:* A clinically compatible case as reported by the patient or healthcare provider, absence of a more likely explanation and one or more of the following laboratory criteria:

- Isolation of chikungunya virus from or demonstration of specific arboviral or genomic sequences in tissue, blood, cerebrospinal fluid (CSF), or other body fluid by polymerase chain reaction (PCR) test (= 5 days after illness onset), immunofluorescence or immunohistochemistry, **or**
- Demonstration of a  $>4$ -fold rise in reciprocal Immunoglobulin G (IgG) antibody titer or Hemagglutination inhibition titer to chikungunya virus antigens in paired acute and convalescent serum samples, **or**
- Demonstration of a  $>4$ -fold rise in PRNT (Plaque reduction neutralization test) end point titer (as expressed by the reciprocal of the last serum dilution showing a 90% reduction in plaque counts compared to the virus infected control) between chikungunya virus and other arboviruses tested in a convalescent serum sample.

*Presumptive/Probable:* A clinically compatible case as reported by the patient or healthcare provider, absence of a more likely explanation and one or more of the following laboratory criteria:

- A positive chikungunya-specific Enzyme-linked immunosorbent assay (ELISA) or immunofluorescence assay (IFA) for immunoglobulin (Ig) M on a single acute or convalescent phase serum specimen.

## **Comment**

**Rule out Dengue Testing.** The differential diagnosis of chikungunya virus infection varies based on place of residence, travel history, and exposures. Dengue and chikungunya viruses are transmitted by the same mosquitoes and have similar clinical features. The two viruses can circulate in the same area and can cause occasional co-infections in the same patient. Chikungunya virus infection is more likely to cause high fever, severe arthralgia, arthritis, rash, and lymphopenia, while dengue virus infection is more likely to cause neutropenia, thrombocytopenia, hemorrhage, shock, and death. It is important to rule out dengue virus infection because proper clinical management of dengue can improve outcome.

## **Chikungunya Reporting**

All infections, regardless of status (i.e., suspect, probable, or confirmed) should be reported using the real-time, secure web-based California Reportable Disease Information Exchange (CalREDIE) system maintained by CDPH. Non-participating jurisdictions should report chikungunya infections by submitting the paper chikungunya case report form by secure email or fax immediately after the investigation is complete. For cases in which no travel history is indicated or local transmission is suspected, CDPH should be notified immediately by telephone.

## **Chikungunya Laboratory Testing**

Serologic (IgG and IgM) and molecular assays to detect evidence of chikungunya virus infection are available through commercial laboratories, from many local public health laboratories, as well as from the CDPH Viral and Rickettsial Disease Laboratory (VRDL). Testing may include:

- *EIA or IFA for IgM and IgG antibodies.* Serologic assays may be cross-reactive with other alphaviruses. When a positive detection is made for chikungunya, VRDL can perform a plaque reduction neutralization assay (PRNT) to attempt to distinguish between chikungunya and other endemic alphaviruses (i.e., western equine encephalitis virus).
- *Real-time RT-PCR (RT-qPCR) for acute serum, plasma, or blood specimens.* Blood for RT-qPCR should be collected within 14 days of symptom onset.

Acute samples that test positive for chikungunya-virus specific antibodies at commercial laboratories should prompt ordering of convalescent testing. Both acute and convalescent samples should be forwarded to VRDL for confirmatory testing.



Samples may be submitted to VRDL using the *General Purpose Specimen Submittal Form* available on the [VRDL Specimen Shipping Guidelines and Current Specimen Submittal Forms webpage](#). Details on orderable tests and specimen requirements are available in the [VRDL Laboratory Test Catalog](#).

**VRDL Contact Information**

Main Telephone Number	(510) 307-8585
Fax Number	(510) 307-8599

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Viral and Rickettsial Disease Laboratory  
Attn: Specimen Receiving  
850 Marina Bay Parkway  
Richmond, CA 94804

## Appendix E

### Zika Surveillance Case Definition, Reporting, and Laboratory Testing

#### Clinical Description (non-congenital Zika virus disease)

To meet the clinical criteria for non-congenital Zika virus disease, the person should have one or more of the following not explained by another etiology:

- Acute onset of one or more of the following symptoms: fever (measured or reported), generalized rash, arthralgia, or non-purulent conjunctivitis,
- Guillain-Barré syndrome,
- Loss of a fetus at greater or equal to 20 weeks gestation.

#### Clinical Description (congenital Zika virus disease)

To meet the clinical criteria for congenital Zika virus disease, the liveborn infant must not have an identified genetic or other cause for the findings, including a positive test for another likely etiology, and should have one or more of the following brain or eye anomalies or neurological sequelae specific for congenital Zika virus disease and typically identifiable in the neonatal period:

- Microcephaly (occipital frontal circumference  $>2$  standard deviations below the mean for age and sex) at birth or postnatal onset,
- Cortical hypoplasia or abnormal gyral patterns (polymicrogyria, lissencephaly, heterotopia),
- Increased volume of cerebrospinal fluid (CSF) (hydrocephalus ex vacuo, unspecified hydrocephalus, ventriculomegaly) due to loss of brain parenchyma,
- Intracranial calcifications (most commonly between the cortex and subcortex),
- Congenital contractures of major joints (arthrogryposis) associated with structural brain anomalies,
- Congenital paralysis of the diaphragm associated with structural brain anomalies,
- Corpus callosum agenesis/hypoplasia,
- Cerebellar hypoplasia,
- Scarring of the macula with coarse deposits of pigment in the retina (focal retinal pigmentary mottling),
- Other structural eye anomalies (microphthalmia, cataracts, chorioretinal atrophy, optic nerve hypoplasia).

#### Laboratory Criteria for Classification (non-congenital Zika virus disease)

##### *Confirmatory laboratory evidence*

- Detection of Zika virus, viral antigen, or viral RNA in a body fluid or tissue; **or**
- Detection of anti-Zika virus IgM antibodies in blood or CSF, with positive Zika virus-specific neutralizing antibody titers and negative neutralizing antibody titers against dengue or other flaviviruses endemic to the region where exposure occurred

#### *Presumptive laboratory evidence*

- Detection of anti-Zika virus IgM antibodies in blood or CSF with a negative anti-dengue virus IgM antibody test in the same specimen with no neutralizing antibody testing performed; **or**
- Four-fold or greater rise in anti-Zika virus-specific neutralizing antibody titers in paired blood specimens; **or**
- In the setting of a Zika virus outbreak with minimal circulation of other endemic flaviviruses, detection of anti-Zika virus IgM antibodies in blood or CSF.

#### **Laboratory Criteria for Classification (congenital Zika virus disease)**

##### *Confirmatory laboratory evidence*

- Detection of Zika virus, viral antigen, or viral RNA in infant CSF, blood, urine, or postmortem tissue; **or**
- Detection of anti-Zika virus IgM antibodies in infant CSF or blood, with positive anti-Zika virus-specific neutralizing antibody titers.

##### *Presumptive laboratory evidence*

- Detection of Zika virus, viral antigen, or viral RNA in amniotic fluid, placenta, umbilical cord, or cord blood \*\* ; **or**
- Detection of anti-Zika virus IgM antibodies in infant CSF or blood \*\* with no neutralizing antibody testing performed.

\*\* To prevent misclassifying postnatal Zika virus infections as congenital cases, in Zika virus endemic areas specimens should be collected within 4 weeks after birth.

#### **Epidemiologic Linkage Criteria**

- Resided in or traveled to an area with a risk of Zika virus transmission in the 14 days before the onset of symptoms, in the 28 days before the onset of Guillain-Barré syndrome, or during pregnancy; **or**
- Laboratory exposure to Zika virus before onset of symptoms or during pregnancy; **or**
- Receipt of blood, blood products, organ transplant, or tissue transplant within 30 days of symptom onset or during pregnancy from a person who has either been diagnosed with Zika virus infection or returned from traveling to an area with risk of Zika virus transmission; **or**
- Sexual contact, within 14 days of symptom onset or during pregnancy, with a person who in the last 90 days has either been diagnosed with Zika virus infection or has returned from traveling to an area with a risk of Zika virus transmission.

#### **Case Classifications (non-congenital Zika virus disease)**

##### *Confirmed*

- Meets the epidemiologic linkage criteria, and clinical and confirmatory laboratory criteria for non-congenital Zika virus disease.

### *Probable*

- Meets the epidemiologic linkage criteria, and clinical and presumptive laboratory criteria for non-congenital Zika virus disease.

## **Case Classifications (congenital Zika virus disease)**

### *Confirmed*

- Meets the clinical criteria for congenital Zika virus disease, **and**
- Meets confirmatory laboratory criteria for congenital Zika virus disease, **and**
- Whose gestational parent meets:
  - Epidemiologic linkage criteria, **or**
  - Confirmatory laboratory criteria for non-congenital Zika virus disease during this pregnancy.

### *Probable*

- Meets the clinical criteria for congenital Zika virus disease, **and**
- Meets presumptive laboratory criteria for congenital Zika virus disease, **and**
- Whose gestational parent meets:
  - Epidemiologic linkage criteria, **or**
  - Confirmatory laboratory criteria for non-congenital Zika virus disease during this pregnancy.

## **Comment**

**Rule Out Dengue Testing.** The differential diagnosis of Zika virus infection varies based on place of residence, travel history, and exposures. Zika, dengue and chikungunya viruses are transmitted by the same mosquitoes and have similar clinical features. These three viruses can circulate in the same area and can cause occasional co-infections in the same patient. Zika virus is more likely to cause fever with maculopapular rash, arthralgia, or conjunctivitis, chikungunya virus infection is more likely to cause high fever, severe arthralgia, arthritis, rash, and lymphopenia, while dengue virus infection is more likely to cause neutropenia, thrombocytopenia, hemorrhage, shock, and death. It is important to rule out dengue virus infection because proper clinical management of dengue can improve outcome.

## **Zika Reporting**

All infections, regardless of status (i.e., probable or confirmed) should be reported using the real-time, secure web-based California Reportable Disease Information Exchange (CalREDIE) system maintained by CDPH on a daily basis. Non-participating jurisdictions should report Zika infections by submitting the paper Zika case report form by secure email or fax immediately after the investigation is complete. For cases in which no travel history is indicated or local transmission is suspected, CDPH should be notified immediately by telephone.

## **Zika Laboratory Testing**

Serologic (IgM) and molecular assays to detect evidence of Zika virus infection are available through commercial laboratories, from many local public health laboratories, as well as from the California Department of Public Health, Viral and Rickettsial Disease Laboratory (VRDL). Any diagnostic test performed for the detection of Zika virus in clinical samples must be granted Emergency Use Authorization (EUA) by the U.S. Food and Drug Administration. A list of current EUA-approved Zika virus assays can be found on their [Emergency Use Authorizations for Medical Devices webpage](#).

Testing may include:

- *EIA, MAC ELISA or immunochromatographic assay to detect IgM antibodies.* Serologic assays may be cross-reactive with other flaviviruses. When a positive detection is made in a Zika virus IgM assay, VRDL can perform a plaque reduction neutralization assay (PRNT) to attempt to distinguish between Zika virus and other exotic (e.g., dengue virus) or endemic (i.e., West Nile virus, St. Louis encephalitis virus) flaviviruses.
- Depending upon the specific assay used, real-time RT-PCR may be performed on serum, whole blood, urine, CSF, or other fluid specimens.

Samples may be submitted to VRDL using the *General Purpose Specimen Submittal Form* available on the [VRDL Specimen Shipping Guidelines and Current Specimen Submittal Forms webpage](#). Details on orderable tests and specimen requirements are available in the [VRDL Laboratory Test Catalog](#).

### **VRDL Contact Information**

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## Appendix F

### Procedures for Processing Mosquitoes for Arbovirus Detection

1. Collect mosquitoes alive and return them immediately to the laboratory. Collections should be kept humid during transport with moist toweling to prevent desiccation. Females held overnight or longer before processing should be offered 5-10% sucrose.
2. Anesthetize mosquitoes by cold, carbon dioxide, or triethylamine (TEA). TEA is recommended because specimens are permanently immobilized with minimal mortality and with no loss of virus titer. TEA should be used either outdoors or under a chemical hood. Collections can be anesthetized outdoors using a few drops of TEA, the specimens transferred to Petri dishes, and then taken into the laboratory for processing. If refrigerated and kept humid, mosquitoes will remain alive in covered Petri dishes for one or two days without additional anesthesia. If mosquitoes are frozen before processing, counting and sorting to species must be done on a chill table to prevent virus loss.
3. Sort mosquito collections to species under a dissecting microscope at 10X to ensure correct identification and to make sure that extraneous mosquito body parts (i.e., legs, wings) or other small insects such as chironomids or *Culicoides* are not inadvertently included in the pools. Avoiding sample contamination is extremely important because diagnostic testing involves highly sensitive RT-qPCR that can detect even very small quantities of virus. Include dead and dried mosquitoes in counts for abundance purposes but exclude them from samples for virus testing. Pools are comprised of 1 to 50 females of each mosquito species from each collection site counted into individual polystyrene vials with snap caps (SPEX Sample Prep #3116) containing two 5mm glass beads. Vials with pools should be labeled sequentially starting with #1 each year after the agency code; e.g., KERN-1-25 to indicate pool #1 for the Kern agency for the year 2025. The same number series should be maintained for all pools, including both invasive *Aedes* and *Culex* species. Data on each pool should be entered online in electronic format through the California Vectorborne Disease Surveillance Gateway ([CalSurv Gateway](https://gateway.vectorsurv.org/)) (<https://gateway.vectorsurv.org/>). Pools to be tested for chikungunya, dengue, and Zika viruses should be marked for this testing in the CalSurv Gateway at the time pools are submitted. **Pools must be accompanied by the “Mosquito Pools Submitted Form MBVS-3” CalSurv form and can only be tested from surveillance sites with documented locations.** Surveillance sites should be registered online at CalSurv Gateway. Pools from unregistered sites (e.g., from door-to-door collections or single-use trap locations) should be assigned the site code “000000” and the exact location should be recorded for each pool using the CalSurv Gateway’s online map.
4. Freeze pools immediately at -80C either on dry ice in an insulated container or in an ultra-low temperature freezer. Pools should be shipped overnight frozen on dry ice or blue ice (minimum 8 blue ice packs) to the Davis Arbovirus Research and Training (DART) laboratory for testing by real-time multiplex RT-PCR. Care must be taken not to allow pools to defrost during storage or shipment because each freeze-thaw cycle may result in a decrease in viral titer; all virus will be lost if the specimens sit at room temperature for extended periods. Agencies will receive an automated email notification when samples are received and when results have been entered into the CalSurv Gateway; additionally,

positive pools will be reported weekly in the California Arbovirus Surveillance Bulletin. If testing for chikungunya, dengue, and Zika viruses is desired, this should be indicated by checking the box for “CDZ Testing” when preparing the online pool submission form in the CalSurv Gateway. Pools can be tested for other *Aedes*-borne viruses such as yellow fever on request.

Ship mosquito pools to the following address:

**When shipping via GLS**

ATTN: Anil Singapuri  
UC Davis, Dept. of PMI  
1285 Veterinary Medicine Mall  
Building: Vet Med 3A, Room 3336  
Davis, CA 95617

**When shipping via FedEx or UPS**

ATTN: Anil Singapuri  
Vet Med 3A, Room 3336  
1285 Veterinary Medicine Mall  
University of California, Davis  
Davis, CA 95616

## Appendix G

### Additional Resources

#### Peer-Reviewed Documents for Vector Control

Amos, B.A., and R.T. Cardé. 2022. Verifying the efficiency of the Biogents Sentinel trap in the field and investigating microclimatic influences on responding *Aedes aegypti* behavior. J. Vector Ecol. 47(2): 166-170. doi: 10.52707/1081-1710-47.2.166

Amos, B.A., S.A. Ritchie, and R.T. Cardé. 2020. Attraction versus capture II: efficiency of the BG-Sentinel trap under semifield conditions and characterizing response behaviors of male *Aedes aegypti* (Diptera: Culicidae). J. Med. Entomol. 57(5): 1539-1549. doi: 10.1093/jme/tjaa065

Bonizzoni, M., G. Gasperi, X. Chen, and A.A. James. 2013. The invasive mosquito species *Aedes albopictus*: current knowledge and future perspectives. Trends Parasitol. 29(9): 460-468. doi: 10.1016/j.pt.2013.07.003

Crepeau, T.N, S.P Healy, K. Bartlett-Healy, I. Unlu, A. Farajollahi, and D.M Fonseca. 2013. Effects of Biogents Sentinel trap field placement on capture rates of adult Asian tiger mosquitoes, *Aedes albopictus*. PLoS ONE 8(3): e60524. doi:10.1371/journal.pone.0060524

Eisen, L., and C.G. Moore. 2013. *Aedes (Stegomyia) aegypti* in the continental United States: a vector at the cool margin of its geographic range. J. Med. Entomol. 50(3): 467-478. doi: 10.1603/ME12245

Farajollahi, A., and D.C. Price. 2013. A rapid identification guide for larvae of the most common North American container-inhabiting *Aedes* species of medical importance. J. Am. Mosq. Control Assoc. 29(3): 203-221. doi: 10.2987/11-6198R.1

Farajollahi, A, S.P. Healy, I. Unlu, R. Gaugler, and D.M. Fonseca. 2012. Effectiveness of ultra-low volume nighttime applications of an adulticide against diurnal *Aedes albopictus*, a critical vector of dengue and chikungunya viruses. PLoS ONE 7(11): e49181. doi: 10.1371/journal.pone.0049181

Fonseca, D.M., I. Unlu, T. Crepeau, A. Farajollahi, S.P. Healy, K. Bartlett-Healy, D. Strickman, R. Gaugler, G. Hamilton, D. Kline, and G.G. Clark. 2012. Area-wide management of *Aedes albopictus*. Part 2: Gauging the efficacy of traditional integrated pest control measures against urban container mosquitoes. Pest Management Science. 69(12): 1351-1361. doi 10.1002/ps.3511

Hawley, W.A. 1988. The biology of *Aedes albopictus*. J. Am. Mosq. Control Assoc. 4: 1-39.

Mackay, A.J., M. Amador, and R. Barrera. 2013. An improved autocidal gravid ovitrap for the control and surveillance of *Aedes aegypti*. Parasites & Vectors 6: 225. doi: 10.1186/1756-3305-6-225



Metzger, M.E., M.H. Yoshimizu, K.A. Padgett, R. Hu, and V.L. Kramer. 2017. Detection and establishment of *Aedes aegypti* and *Aedes albopictus* (Diptera: Culicidae) mosquitoes in California, 2011-2015. J. Med. Entomol. 54(3): 533-543. doi: 10.1093/jme/tjw237

Unlu, I., A. Farajollahi, D. Strickman, D.M. Fonseca. 2013. Crouching tiger, hidden trouble: urban sources of *Aedes albopictus* (Diptera: Culicidae) refractory to source-reduction. PLoS ONE 8(10): e77999. doi: 10.1371/journal.pone.0077999

### Peer-Reviewed Documents for Public Health Lessons

Adalja A.A., T.K. Sell, N. Bouri, and C. Franco. 2012. Lessons learned during dengue outbreaks in the United States, 2001-2011. Emerg. Infect. Dis. 18(4): 608-14. doi: 10.3201/eid1804.110968

Effler P., L. Pang, P. Kitsutani, V. Vorndam, M. Nakata, T. Ayers, J. Elm, T. Tom, P. Reiter, J.G. Rigau-Perez, J.M. Hayes, K. Mills, M. Napier, G.G. Clark, D.J. Gubler, and the Hawaii Dengue Outbreak Investigation Team. 2005. Dengue fever, Hawaii, 2001-2002. Emerg. Infect. Dis. 11(5): 742-9. doi: 10.3201/eid1105.041063

Feaster, M., R. Patrick, M. Oshiro, M. Kuan, Y-Y. Goh, M. Carmona, S.Y. Tartof, J. Farned, T. Hallum, A.J. Lund, C. Preas, S. Messenger, V. Kramer, M. Danforth, and C. Sheridan. Notes from the field: first locally acquired dengue virus infections – Pasadena, California, October-December 2023. MMWR Morb Mortal Wkly Rep. 2024 Oct 24; 73(42): 955-56. doi: 10.15585/mmwr.mm7342a4

Locally acquired Dengue--Key West, Florida, 2009-2010. Centers for Disease Control and Prevention (CDC). MMWR Morb Mortal Wkly Rep. 2010 May 21; 59(19): 577-81. <http://www.cdc.gov/mmwr/preview/mmwrhtml/mm5919a1.htm>

Porse, C.C., V. Kramer, M.H. Yoshimizu, M. Metzger, R. Hu, K. Padgett, and D. Vugia. 2015. Public health response to *Aedes aegypti* and *Ae. albopictus* mosquitoes invading California, USA. Emerg. Infect. Dis. 21(10): 1827-1829. doi: 10.3201/eid2110.150494

Porse C.C., S. Messenger, D.J. Vugia, W. Jilek, M. Salas, J. Watt, and V. Kramer. 2018.. Travel-associated Zika cases and threat of local transmission during global outbreak, California, USA. Emerg. Infect. Dis. 24(9): 1626-1632. doi: 10.3201/eid2409.180203

Rodriguez D.M., Z.J. Madewell, J.M. Torres, A. Rivera, J.M. Wong, G.A. Santiago, V. Rivera-Amill, G. Paz-Bailey, M. Marzan-Rodriguez, and L.E. Adams. 2024. Epidemiology of Dengue — Puerto Rico, 2010–2024. MMWR Morb Mortal Wkly Rep. 73(49): 1112–1117. doi: 10.15585/mmwr.mm7349a1

## Surveillance and Control Manuals

American Mosquito Control Association. Best Practices for Integrated Mosquito Management: A Focused Update. January 2017.

[https://www.mosquito.org/assets/pdf/amca\\_guidelines\\_final\\_pdf/](https://www.mosquito.org/assets/pdf/amca_guidelines_final_pdf/)

Centers for Disease Control and Prevention. Surveillance and Control of *Aedes aegypti* and *Aedes albopictus* in the United States.

<https://www.cdc.gov/mosquitoes/pdfs/mosquito-control-508.pdf>

European Centre for Disease Prevention and Control. Guidelines for the surveillance of invasive mosquitoes in Europe. Stockholm: ECDC; 2012.

<https://ecdc.europa.eu/sites/portal/files/media/en/publications/Publications/TER-Mosquito-surveillance-guidelines.pdf>

Florida Department of Health. Surveillance and Control of selected Mosquito-Borne Disease in Florida: 2014 Guidebook. [http://www.floridahealth.gov/diseases-and-conditions/mosquito-borne-diseases/\\_documents/arboguide-2014.pdf](http://www.floridahealth.gov/diseases-and-conditions/mosquito-borne-diseases/_documents/arboguide-2014.pdf)

World Health Organization: Dengue Guidelines for Diagnosis, Treatment, Prevention, and Control, 2009. <https://iris.who.int/handle/10665/44188>

## General Resources

CDPH Vector-Borne Disease Section. Includes links to dengue, chikungunya, and Zika webpages <https://www.cdph.ca.gov/Programs/CID/DCDC/Pages/VBDS.aspx>

CDPH *Aedes aegypti* and *Aedes albopictus* Mosquitoes

<https://www.cdph.ca.gov/Programs/CID/DCDC/Pages/Aedes-aegypti-and-Aedes-albopictus-mosquitoes.aspx>

CDPH Information for Clinicians: *Aedes aegypti* and *Aedes albopictus* Mosquitoes in California and Reporting Patients with Suspected Dengue or Chikungunya to Public Health

<https://www.cdph.ca.gov/Programs/CID/DCDC/CDPH%20Document%20Library/DengueorChikungunyaInformationForCliniciansinCA.pdf>

CDPH Selected traps and collection methods for *Aedes aegypti* and *Aedes albopictus* surveillance and control

<https://www.cdph.ca.gov/Programs/CID/DCDC/CDPH%20Document%20Library/CDPHAedesTrapSummaries.pdf>

CDC Chikungunya Information. <https://www.cdc.gov/chikungunya/>

CDC Dengue Information. <https://www.cdc.gov/dengue/>

CDC Zika Information. <https://www.cdc.gov/zika/>

CDC - Mosquito Surveillance Traps

<https://www.cdc.gov/mosquitoes/php/toolkit/mosquito-surveillance-traps.html>

VectorSurv invasive *Aedes* maps. <https://maps.vectorsurv.org/invasive/>