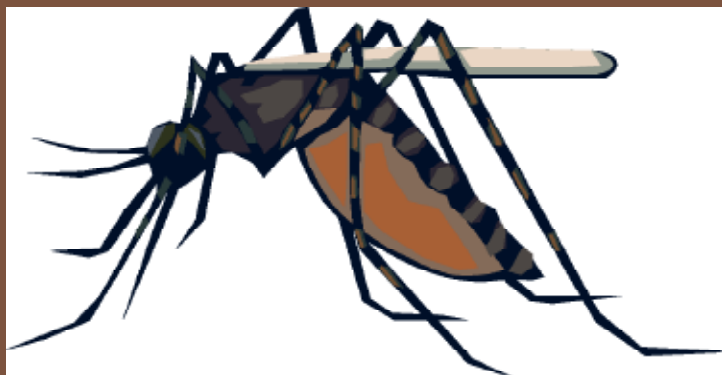


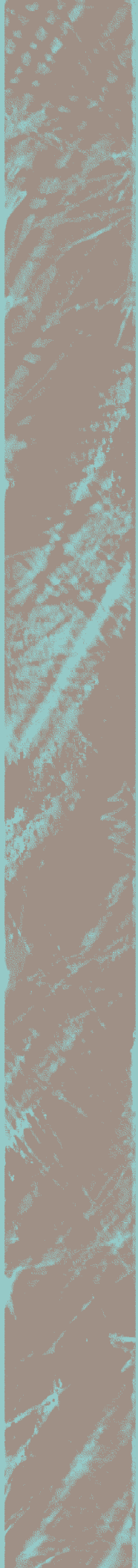
Guidance for Surveillance, Prevention, and Control of Mosquito-borne Disease

2008 Edition



Prevention through Partnerships

Washington State Department of Health
Zoonotic Disease Program



Guidance for Surveillance, Prevention, and Control of Mosquito- borne Disease

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Overview

Purpose

This guidance document provides background information and establishes procedures for state and local agencies in preparing for and responding to the presence of mosquito-borne viruses (included in the category of arboviruses [**arthropod-borne viruses**]) and the illnesses they cause. Detection of certain arboviruses, such as western equine encephalitis virus, St. Louis encephalitis virus, or West Nile virus in mosquitoes or animal populations requires prompt action to reduce the risk of human infection. The detection of mosquito-borne infections in humans or animals may require additional control activities. This guidance sets forth recommended surveillance, prevention, and response procedures and describes state and local roles in responding to mosquito-borne disease.

The appendices include detailed information on basic mosquito biology and habitat, surveillance techniques, guidelines for specimen collection for birds, horses, and humans, and mosquito control procedures. Also provided are model press releases, key contacts, and statutes on mosquito control and formation of mosquito control districts.

Background

Washington State has a history of outbreaks of mosquito-borne diseases such as western equine encephalitis and St. Louis encephalitis. The earliest recognized outbreak occurred in central and eastern Washington during the summers of 1939-1942 when approximately 200 people were infected with a mosquito-borne disease in Yakima, Chelan, Okanogan, and Whitman counties. Hundreds, perhaps thousands, of horses developed encephalomyelitis during this period. Research and surveillance projects identified *Culex tarsalis* as the primary mosquito vector in the Yakima Valley area. More information about mosquito-borne diseases is available in Appendix D.

Both human and animal infections occurred sporadically in the following decades. These mosquito-borne disease outbreaks were the driving force for creation of many of the currently operating mosquito control districts in Washington. Sixteen mosquito control districts now operate in 14 counties. Twelve of the districts are in eastern Washington. Districts play a major role in identifying mosquito larval habitats, conducting larval and adult control activities, and educating the public on bite prevention and habitat reduction.

Since the establishment of many of the early mosquito control districts in Washington, non-human surveillance for mosquito-borne disease has been sporadic. However, the outbreak of West Nile virus (WNV) encephalitis in the

New York City metropolitan area during the summer of 1999 and the virus' continued geographic expansion in the following years has resulted in increased surveillance activity across the United States. Washington State Department of Health (DOH) and its partners initiated activities to improve mosquito-borne disease surveillance in 2000. DOH, under a cooperative agreement from the Centers for Disease Control and Prevention (CDC), began working to enhance routine surveillance for human mosquito-borne diseases and to establish routine surveillance for infected mosquitoes and animals with local health jurisdictions, mosquito control districts, military installations, and other partners. The cooperative agreement supported development of enhanced disease surveillance systems for humans and horses in cooperation with physicians, veterinarians, and local health jurisdictions. A dead bird surveillance network was established for collection and testing of the most susceptible birds, primarily Corvids (crows, ravens, jays, and magpies), for WNV. Mosquito surveillance activities were initiated for the purpose of identification and mosquito pool testing. See Appendix D for a summary of WNV data collected since the establishment of the WNV program in 2000. In 2006, with the continued establishment of WNV in new areas of the state, Washington detected the first human infections acquired in Washington (3) and an increase in environmental detections (13 birds and 6 horses). Fewer environmental detections (8 horses, 1 dog, and 1 bird) were confirmed in 2007, but all horse detections were tightly clustered in the same area in Yakima County.



Surveillance

Mosquito Surveillance

Mosquito surveillance is an essential component of a comprehensive mosquito-borne disease prevention and control program. The objective of mosquito surveillance is to determine species composition, geographic distribution, seasonal occurrences, and abundance of potential vectors of mosquito-borne pathogens within each county by collecting and identifying larval and adult mosquitoes. Up-to-date information on mosquito species and their distribution is essential to developing effective prevention and control programs. Samples of adult female mosquitoes can be pooled by species and tested for the presence of mosquito-borne viruses, which helps determine potential enzoonotic and bridge vector species in an area.

Components of an effective program include: identifying and mapping larval mosquito habitats using ground-based and aerial surveillance methods, identifying and mapping the location of potential vector species within each county through the collection of adult or larval mosquitoes, and testing of potential vector species for mosquito-borne viruses.

Mosquito surveillance should typically begin in April or May and continue through September or into October, depending on weather conditions. Collections should be made in a variety of habitats throughout the season. Information on basic mosquito biology and common habitats in Washington is included in Appendix A. Procedures for larval and adult surveillance are discussed in Appendix B. DOH's Zoonotic Disease Program and its partners are available to provide training on these topics.

Bird and Mammal Surveillance

Birds and mammals can be important sentinels for mosquito-borne viruses and may provide early warning to allow for control actions to prevent human cases and reduce impacts on livestock, pets, and wildlife. Avian morbidity and mortality surveillance appears to be one of the most sensitive early detection system for WNV and should be a component of every mosquito-borne disease surveillance program. The surveillance system should include two components:

- a reporting system for sightings of dead or ill birds to track increases possibly due to WNV.
- submission of selected individual birds for testing.

The effectiveness of dead or ill bird reporting will be enhanced by collaboration with groups and individuals most likely to find dead or ill birds, such as agencies

whose employees spend considerable time outdoors (parks, fish and wildlife, public utilities) and members of birding and outdoor recreational organizations.

Avian surveillance should be initiated when local adult mosquito activity begins in the spring. A database should be established to record and analyze dead bird sightings with the following information: caller identification, date observed, location, species, and condition. This information should be reviewed periodically to detect increases in avian mortality. Birds chosen for testing should have died within the last 48 hours and be in good condition with no decomposition or severe trauma. Birds from areas showing a general increase in observed bird deaths are of particular value. Testing of corvids should be emphasized, as they are very susceptible to WNV infection. Raptor species (hawks, owls, eagles, and vultures) are also susceptible and should be considered for testing. More information on bird surveillance and submission protocols is in Appendix C. Collection and shipping materials are available from DOH.

Live sentinel animals can also be used for mosquito-borne virus surveillance. Several species of birds, both wild and domestic, have been used for surveillance programs. Sentinel chickens have proven to be an important tool associated with monitoring western equine encephalitis (WEE) and St. Louis encephalitis (SLE) in the western United States. Although sentinel chickens are often not the first indication of WNV in an area, they do provide evidence of local transmission. Chickens are easy to handle and have been used in Washington for detection of western equine encephalitis, St. Louis encephalitis, and WNV in the environment. Caged chickens are placed in areas of known mosquito activity and blood samples from the birds are periodically tested for antibodies to the mosquito-borne viruses of concern. Positive results indicate an infected mosquito had bitten them. This information can help in determining appropriate response activities when used as part of a comprehensive surveillance program. Mosquito control districts or other agencies and organizations with a field capability and laboratory support can effectively operate sentinel chicken flock programs. DOH and mosquito control districts can provide information on establishing a sentinel flock program.

Domestic animals, horses, and poultry, can also provide information about mosquito-borne viruses in a community. Surveillance by testing ill domestic animals is likely to be the least sensitive and specific because most animals that become ill with clinical signs of neurological disease are likely to have an illness other than a mosquito-borne disease. However, testing of symptomatic horses for WEE and WNV is appropriate. Unvaccinated horses have been found to be particularly susceptible to WNV, with an estimated one-third of those becoming ill, dying. Veterinarians should report equine neurological disease of unknown origin to the Washington State Department of Agriculture at 360.902.1878. Department of Agriculture personnel will consult with the reporting veterinarian

to determine if the animal should be tested for mosquito-borne virus or for other neurological conditions such as rabies. More equine surveillance and vaccine information is included in Appendix C. Also, poultry testing after sentinel flocks show positive antibody response can help identify geographical spread of WEE, SLE, and WNV. Only persons trained in obtaining blood samples from domestic poultry should be involved in such efforts.

Human Surveillance

Prevention and control of human disease is the goal of surveillance for mosquito-borne diseases. Due to historical outbreaks of mosquito-borne disease in the Yakima Valley, surveillance for human arboviral infection has been long-standing in Washington State. Healthcare providers are often on the frontline in the detection of human disease outbreaks, and we rely on them for rapid reporting of suspected notifiable conditions, including arboviral infections. Surveillance for human arboviral disease is a year-round activity; however, during months when mosquito activity is greatest, local health jurisdictions typically alert their healthcare community to be on alert for potential WNV infections. Additional information about reporting and investigating suspected human arboviral infections can be found at <http://www.doh.wa.gov/notify/nc/arbo.htm>.

Confidentiality of Surveillance Information

The sharing of health information between healthcare providers and public health investigators is routine in the prevention and control of notifiable conditions. Information shared with the public is aggregate – pooled data without individual identifiers. Individuals are never publicly identified because disclosure of their identity violates Washington State regulations that guarantee the protection of an individual's health-related information. The rapid sharing of animal and mosquito surveillance results with agencies and the public is essential for development of appropriate disease control measures.

In addition to the protection of human identifiable information, protection of confidentiality is important for bird and mammal information. By policy, DOH does not initiate disclosure of information that identifies the animal owner's name and street address, veterinarian's name and address, name and address of individual or institution submitting a specimen, and in some cases, species of bird or mammal. However, in responding to a public disclosure request, no state or federal law prohibits DOH from disclosing specific information on diagnostic testing of animals or mosquitoes.



Prevention and Public Information

Enhanced public awareness and prevention education are two important components of an effective mosquito-borne disease prevention and control program. Outreach strategies can promote public cooperation in reducing man-made collections of standing water in which mosquitoes develop. They can also help individuals reduce their risk of being bitten by promoting bite prevention techniques. Health care providers can also be informed about the diagnosis and treatment of human arboviral encephalitis.

A public health education campaign about mosquitoes and mosquito-borne diseases should accomplish the following objectives:

- Encourage personal protection techniques, such as restricting outdoor activities when mosquitoes are active (dusk to dawn), wearing long-sleeved shirts and long pants, and using an effective mosquito repellent that contains an active ingredient that has been registered with the U.S. Environmental Protection Agency (EPA).
- Inform the public about how to reduce mosquito habitat by eliminating stagnant water on privately owned property.
- Improve public understanding of the mosquito-borne disease cycle and its sources and reservoirs in mosquitoes, birds, and mammals.
- Increase awareness among the public and health professionals of the potential risk for infection with mosquito-borne disease locally and when traveling to other areas.
- Encourage health care providers to promptly report cases of human encephalitis.
- Improve knowledge among health care providers of the signs and symptoms of human arboviral encephalitis.

Public messages tell the community about agency activities and provide suggestions on property maintenance and personal protection. Requests for public help in observation and reporting of dead birds tend to increase participation and awareness. Suggested timing of messages on mosquitoes and mosquito-borne disease for the public along with example news releases on WNV are presented in Appendix F. Messages are organized according to virus activity. Each builds upon preceding messages by reinforcing earlier messages.



Mosquito Control

Prevention and control of mosquito-borne diseases is accomplished most effectively through a comprehensive, integrated pest management approach. Such a program is effective at pest control and minimizes public exposure to substances used for control. Programs are not intended to entirely eliminate mosquitoes, but rather reduce their numbers and therefore reduce the risk of disease transmission.

Mosquito control professionals use several methods to reduce mosquito populations. These include reducing the available habitat for larval mosquito development, control of mosquito larvae, and control of adult mosquito populations. Mosquito control professionals can also engage in public education campaigns to encourage property owners to reduce of mosquito habitat on privately owned property and encourage personal protection measures to avoid mosquito bites.

Habitat reduction no longer involves extensive changes to the natural environment. Years ago, filling or draining natural wetlands was practiced in an effort to reduce mosquito breeding areas. Today, efforts focus on modifying or eliminating larval habitats by water management practices that reduce flooding or minimize standing water in irrigated areas. Eliminating water-filled containers, such as buckets, plant saucers, cans, and tires can also help. Maintaining or draining pools, tubs, water troughs and birdbaths, and cleaning roof gutters are additional steps property owners can take. The use of pumps and fountains in landscape ponds can keep water from stagnating. Fish that prey on mosquito larvae may be added to contained ponds. However, Washington law prohibits importing or releasing certain fish and wildlife species known to have a damaging effect on native species. Contact the Washington Department of Fish and Wildlife before introducing any fish into aquatic environments.

When habitat reduction or water management is not feasible or have failed to adequately control mosquito populations, chemical or biological control may be required. Treatments may be directed at either the immature or adult stage of the mosquito life cycle.

Larviciding, the application of chemicals to kill mosquito larvae or pupae in the water, is generally more effective and target-specific than applying chemicals to kill adult mosquitoes (adulticiding), but less permanent than habitat reduction. The objective of larviciding is to control the immature stages in their aquatic habitat at the developmental habitat before adult populations have had a chance to disperse, and to maintain populations at levels that reduce the risk of mosquito-borne virus transmission. Larvicides can be applied from the ground or by aerial application if large or inaccessible areas must be treated. Several materials in various formulations (briquet, granular, liquid, pellet, powder, water

soluble pouch) are labeled for mosquito larviciding and include the bacterial larvicides *Bacillus thuringiensis israelensis (Bti)* and *Bacillus sphaericus*; methoprene, an insect growth regulator; monomolecular surface films; larvicidal oils, petroleum based and mineral based; and the organophosphates temephos and malathion. Detailed information on larval control compounds is included in Appendix E. Applications of larvicides often encompass fewer acres than adulticides because treatments are made to relatively small areas where larvae are concentrated as opposed to larger regions where adults have dispersed. Selection of larvicide products must be appropriate for the habitat being treated, accurately applied, and based on surveillance data.

Adulticiding is usually the least efficient mosquito control technique. However, it is the only way to kill adult mosquitoes, and is the last line of defense in reducing mosquito populations. Adulticides are typically applied as an Ultra-Low-Volume (ULV) spray where small amounts of insecticide are dispersed either by truck mounted equipment or aircraft. The insecticide must drift through the habitat in which mosquitoes are flying in order to provide optimal control benefits. Barrier treatment, typically applied as high volume liquids with hand held spray equipment using compounds with residual characteristics, is a technique that may provide control benefits along property boundaries that are near mosquito-producing habitats. Adulticides labeled for mosquito control include the organophosphates malathion and naled, some natural pyrethrins, and synthetic pyrethroids (permethrin, resmethrin, and sumithrin). Insecticide selection and time of application should be based on the distribution and behavior of the target mosquito species. Additional information and considerations for using adulticides is included in Appendix E.



Roles and Activities

Local Health Jurisdictions

Local health jurisdictions have the authority to take measures to control and eliminate mosquitoes in order to prevent and control the spread of disease transmitted by mosquitoes (RCWs 70.05.060 and 70.05.070).

Local health jurisdictions may within available resources provide for a routine mosquito-borne disease education, surveillance, and prevention program. Such a program may include larval and adult mosquito surveillance, dead bird surveillance, human and horse case surveillance, and a public awareness and education campaign. Such a program may include the activities described below:

- Development of plans for responding to a mosquito-borne disease outbreak.
- Larval surveillance, which may include the location, mapping, and characterization of mosquito habitats, larval collection of mosquito larvae from these habitats, and submission for identification of species.
- Adult surveillance, which may include collection of adult mosquitoes periodically during the months of April through October, and submission for identification of species and, if possible, for pooling of females to test for mosquito-borne virus.
- Recording of all available jurisdictional data collected from mosquito surveillance.
- Authority to facilitate and coordinate the use of larvicides and adulticides for mosquito control.
- Identification of organizations within the local health jurisdiction with capabilities of conducting larval and adult mosquito control.
- Education of local governing bodies on the creation of mosquito control districts.
- Dead bird surveillance from April through October, with collection of fresh specimens of appropriate species for submission for viral testing.
- Human and horse case surveillance throughout the year in cooperation with physicians and veterinarians.
- Public information and education campaigns from April through the end of mosquito season as appropriate for the level of mosquito and disease activity.

Counties and Municipalities

These governmental entities, under their broad powers to provide for the public health and safety, can generally undertake mosquito control measures on lands and bodies of water under their control, including larviciding and adulticiding, when indicated and with proper licensing and permits (WA Constitution Article XI, Section II; Titles 35, 35A, and 36 RCW).

Counties and municipalities must cooperate with state mosquito control plans (RCW 70.22.060).

Mosquito Control Districts

Mosquito control districts, operating under RCW Chapter 17.28, may take all necessary or proper steps for the extermination of mosquitoes and abate, as nuisances, all stagnant pools of water or other breeding places for mosquitoes with oversight by the county or city in which they exist. Mosquito control districts are funded through tax levies to provide larviciding and adulticiding as appropriate. They also have the power to enter without hindrance upon any land within the district for the purpose of inspection to ascertain whether breeding places of mosquitoes exist upon such lands. In addition to the above powers, mosquito control districts may provide assistance to local health jurisdictions by:

- Collection and identification of mosquitoes.
- Mapping of mosquito habitat and location of disease vector species.
- Participating in dead bird surveillance and collection.
- Working with local health jurisdictions, cities, and counties to facilitate or provide mosquito control, including abatement through larviciding and adulticiding as appropriate.
- Directing private property owners within its district to control mosquitoes on their land, and causing such control to occur at the expense of the property owner under certain circumstances.

Washington State Department of Health

The Department of Health has the authority to conduct studies to determine the effect of mosquitoes as a health hazard (RCW 70.22.020).

DOH will provide: training and technical assistance on mosquito-borne disease surveillance, laboratory services to support surveillance efforts, and mosquito disease prevention messages for public information campaigns. DOH will conduct training for local health, mosquito control districts, and other interested parties. DOH will provide assistance on the following:

- Identification and mapping of mosquito breeding habitat.
- Techniques for surveillance of adult and larval mosquitoes.
- Permit coverage for aquatic mosquito control.
- Dead bird surveillance, collection, and preparation for shipping to the lab.
- Lab testing for dead birds and mosquitoes.
- Data recording and management.
- Public information messages, news releases, brochures, web pages, and a toll-free information line.
- Statewide data on surveillance information collected through local health jurisdictions and other partners.
- Development of plans for responding to a mosquito-borne disease outbreak.

DOH will coordinate plans for mosquito control work that may be projected by a local, state or federal entity and to arrange for cooperation between such entities or individuals. DOH will facilitate cooperation if outbreak conditions are spread across jurisdictions, and cooperation among parties within different jurisdictions is required. During outbreak conditions, the secretary may request that the governor declare a public health emergency (RCW 70.22.030).

- DOH has the authority to conduct larval or adult mosquito control, if funds are available.

Washington State Department of Ecology

The Department of Ecology is required by law to protect the waters of the state from actions that might pollute or harm them. The vehicle for allowing the application of pesticides to waterways is the National Pollution Discharge Elimination System (NPDES) Permit. This permit is administered by the Department of Ecology, and anyone wishing to apply pesticides for aquatic mosquito control into waters of the state must obtain a NPDES permit prior to application.

The Department of Ecology has issued DOH a statewide blanket NPDES permit for aquatic mosquito control. DOH is extending its coverage of the permit to any entity in the state qualified to follow the conditions of the permit and the best management practices for mosquito control.

Washington State Department of Agriculture

The Department of Agriculture regulates the labeling and registration of pesticides used in the state and has authority over the applicators of pesticides and controls their actions through a license. Except for limited homeowner applications of some pesticides available commercially at the retail level, an applicator of larvicides or adulticides must possess an applicator's license. This license mandates reporting requirements and ensures appropriate knowledge and accountability of the license holder for personal safety and environmental protection in the application of materials.

WSDA issues permits for the sale and distribution of veterinary biologics, including fully licensed WNV equine vaccines available through licensed veterinarians. WSDA also enforces the animal disease reporting requirement of veterinarians, veterinary laboratories, and others. WSDA is the lead state agency for WNV in equines.

Washington State Department of Fish and Wildlife

The Department of Fish and Wildlife addresses concerns with potential impacts of WNV on bird populations and effects of mosquito control activities on wildlife populations.

Washington Parks and Recreation Commission

The Washington Parks and Recreation Commission protects the health and safety of park visitors and park employees. The agency may apply appropriate insect management controls.

Washington State Department of Transportation

The Department of Transportation controls larvae of WNV vector mosquitoes located in Department of Transportation rights-of-way.



Guidelines for a Phased Response

The principal goal of a phased response is to minimize the health impacts of mosquito-borne diseases in humans, and in livestock, domestic and zoo animals. Experts have a growing, but still incomplete understanding of the ecology and epidemiology of some mosquito-borne viruses (including WNV) in the United States. The occurrence of mosquito-borne disease is sporadic, and prevention methods have limitations. It is reasonable to assume that prevention and control measures, no matter how intensive, cannot prevent all human mosquito-borne infections. However, surveillance information can be used to estimate the risk to humans, and to develop appropriate response measures. Appropriate and timely response to surveillance data is key to preventing human and animal disease associated with WNV and other mosquito-borne viruses. That response must include effective mosquito control and public education without delay, if an increasing intensity of virus activity is detected by bird- or mosquito-based surveillance systems.

The following table defines six WNV alert levels (ranging from 0 or “No Risk” to 5, “Outbreak in Progress”) and provides guidelines for appropriate surveillance, education, and control strategies for each alert level. The levels may be applied to individual counties or regions of the state. Depending on surveillance findings, areas within counties may be in different alert levels based on surveillance activity, geography, known mosquito habitat, population centers, and other criteria. Local and regional characteristics may alter the alert level at which specific actions must be taken.

Strategies used at the county level will vary, depending on availability of resources. DOH will partner with counties to help maximize use of existing resources.

Flexibility is required when implementing the guidelines for a phased response. Specific and detailed recommendations that will fit all possible scenarios are not possible, especially at a local level. Therefore, public health action should depend on interpreting the best available surveillance data in an area, in light of these general guidelines. In addition, the following factors should be considered when translating these guidelines into a plan of action:

- Current weather and predicted climate anomalies.
- Quality, availability, and timeliness of surveillance data.
- Flexibility of the planned prevention and control activities, given existing budget and infrastructure.
- Public acceptance of the planned prevention and control strategies.

- Expected future duration of WNV transmission (surveillance events earlier in the transmission season will generally have greater significance).
- Other ongoing mosquito control activities, such as nuisance mosquito control or vector mosquito control for established arboviral encephalitis viruses.

Alert Levels for a Phased Response

Alert Level 0

No Risk of Human Outbreak Off-season (winter): adult vectors inactive; climate unsuitable.

Surveillance Response	Education Response	Mosquito Control Response	Communications
Recommended Response for Local Health Jurisdictions and other local entities¹			
<ul style="list-style-type: none"> Analyze environmental surveillance results from previous season. Conduct ongoing surveillance and investigation of human mosquito-borne diseases. Develop West Nile virus response plan in conjunction with community partners. Review and update mosquito and bird surveillance plans for coming season. Secure equipment, materials, funding, and other resources. 	<ul style="list-style-type: none"> Evaluate effectiveness of educational materials and outreach from previous season and update plans for coming season. Develop partnerships with media and work with community leaders, local partners, and organizations on plans for education of the general public through targeted prevention messages. 	<ul style="list-style-type: none"> Secure surveillance and control resources necessary to enable appropriate response. Obtain necessary licenses and permits required for mosquito control. Establish mosquito control contracts with government or private organizations. 	<ul style="list-style-type: none"> Review and update local communications plans for the coming season. Facilitate planning and coordination among local partners, including municipalities, mosquito control districts, and other local and regional agencies and organizations. Alert DOH to gaps in local surveillance, education, and control plans.
Recommended Response for Washington State Department of Health			
<ul style="list-style-type: none"> Coordinate the statewide WNV surveillance program. Secure resources and develop agreements for laboratory testing of specimens. Collate statewide surveillance data and forward to the Centers for Disease Control (CDC). Update statewide protocols for mosquito and dead bird surveillance. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Develop and distribute educational materials. Develop and conduct general presentations that include information on surveillance, prevention, and control. Conduct audience research to develop and target education and community involvement. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Extend coverage of the Aquatic Mosquito Control National Pollutant Discharge Elimination System (NPDES) General Permit to local governments and other qualified entities to control mosquito larvae in waters of the state. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Review and update statewide communications plans for the coming season. Brief state agency partners on the status of mosquito-borne disease in Washington. State agency partners provide updates on WNV response activities. Maintain a website with current information on environmental and human surveillance in Washington. Maintain a toll-free telephone line with information on reported human infections, modes of disease transmission, risks, and preventive measures. DOH provides technical assistance, consultation, and coordination, as needed, for the development of local plans.

¹ Includes municipalities, mosquito control districts, and other local and regional agencies and organizations.

Alert Level 1

Remote Risk of Human Outbreak

Spring, summer, or fall: No detections of mosquito, animal, or human surveillance findings in the current calendar year.

Surveillance Response	Education Response	Mosquito Control Response	Communications
Recommended Response for Local Health Jurisdictions and other local entities			
<ul style="list-style-type: none"> Conduct mosquito surveillance and record and report results. Conduct dead bird surveillance and record and report dead bird activity. Inventory and map surveillance findings and new mosquito habitats. Track occurrence of equine cases. Ongoing surveillance and investigation of human mosquito-borne diseases. 	<ul style="list-style-type: none"> Conduct community outreach and public education emphasizing source reduction and bite prevention. Identify high risk populations (+50 years old, immune compromised, significant outdoor exposure) and locations (outdoor venues used in the evening hours). 	<ul style="list-style-type: none"> Promote source reduction (removal of mosquito breeding habitat) where possible. Follow local response plans that include guidelines for conducting mosquito control using integrated pest management principles. Consider promoting the use of mosquito larvicides at specific locations identified as having potential amplifying and bridge vectors species present and where larval counts meet or exceed an established level. 	<ul style="list-style-type: none"> Alert DOH to gaps in local surveillance, education, and control plans under changing conditions.
Recommended Response for Washington State Department of Health			
<ul style="list-style-type: none"> Coordinate with local partners, laboratories, and other contracted services for state-wide mosquito collection, identification, and testing. Coordinate with local partners, laboratories, and other contracted services for state-wide dead bird surveillance. Provide information to health care providers on recognition, diagnosis, laboratory testing, and reporting of mosquito-borne diseases. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Conduct community outreach and public education emphasizing source reduction and bite prevention. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> DOH provides technical assistance and coordination, as needed, for development and implementation of local plans. Alert local health to existing, updated, and new educational materials available for statewide use. Environmental surveillance partners report unconfirmed environmental positives to the DOH ZD Program. Labs report confirmed environmental positives to the DOH ZD Program. The ZD Program reports confirmed environmental positives to internal and external partners. Positive human test results are reported to DOH Office of Communicable Disease Epidemiology. The Office of Communicable Disease Epidemiology reports human positives to internal and external partners.

Alert Level 2

Low Risk of Human Outbreak

Summer or fall: Limited or sporadic mosquito-borne virus detection in birds and/or mosquitoes prior to August 1.

Surveillance Response	Education Response	Mosquito Control Response	Communications
Recommended Response for Local Health Jurisdictions and other local entities			
<ul style="list-style-type: none"> Continue surveillance activities described in Alert Level 1. Ongoing surveillance and investigation of human mosquito-borne diseases with alerts to health providers as indicated. Conduct mosquito surveillance in areas of positive findings and in adjacent areas to identify water body sources where mosquitoes develop. Identify mosquito species and test for disease. Provide information to veterinarians on local mosquito-borne virus activity and suggest increased vigilance for equine cases. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 1. Increase public education utilizing communication partnerships. Conduct risk communication about mosquito control measures that are or may be taken, including adult mosquito control. Emphasize source reduction. Emphasize personal protection, particularly for persons over 50 years of age, the immunocompromised, and people with significant outdoor exposure. 	<ul style="list-style-type: none"> Continue control activities described in Alert Level 1. Promote expanded larval control and source reduction, including areas adjacent to those with mosquito-borne disease activity. 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 1.
Recommended Response for Washington State Department of Health			
<ul style="list-style-type: none"> Continue coordination of the statewide WNV monitoring system, as described above. With WSDA, provide information to veterinarians on local mosquito-borne virus activity and suggest increased vigilance for equine cases. Continue other surveillance activities described in Alert Level 1. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 1. Increase public education utilizing communication partnerships. Conduct risk communication about mosquito control measures that are or may be taken, including adult mosquito control. Emphasize source reduction. Emphasize personal protection, particularly for person over 50 years of age, the immunocompromised, and people with significant outdoor exposure. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue mosquito control activities describe in Alert Level 1. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 1.

Alert Level 3

Moderate Risk of Human Outbreak

Spring, summer, or fall: Detection of mosquito-borne virus in birds before August 1, or confirmed equine or human infections, or sustained mosquito-borne virus detection in birds or mosquitoes in the absence of human infections.

Surveillance Response	Education Response	Mosquito Control Response	Communications
Recommended Response for Local Health Jurisdictions and other local entities			
<ul style="list-style-type: none"> Continue surveillance activities described in Alert Level 2. Expand mosquito and bird surveillance as necessary. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 2. Expand public information using TV, radio, and newspaper media. Emphasize personal protection and source reduction. Enhance risk communication about adult mosquito control. Engage local leaders and organizations to speak and inform their communities about the mosquito-borne disease. 	<ul style="list-style-type: none"> Continue control activities described in Alert Level 2. Continue larval control efforts. Consider implementing adult control if mosquito populations exceed locally established threshold levels.² Focus control efforts where surveillance indicates potential for human risk to increase. 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 2.
Recommended Response for Washington State Department of Health			
<ul style="list-style-type: none"> Continue coordination of the statewide WNV monitoring system, as described above. Continue other surveillance activities described in Alert Level 2. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 2. Expand public information using TV, radio, and newspaper media. Emphasize personal protection and source reduction. Enhance risk communication about adult mosquito control. Engage local leaders and organizations to speak and inform their communities about the mosquito-borne disease. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue mosquito control activities described in Alert Level 2. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 2. Consider activating the DOH Assessment Response Team (ART).

² In general, the finding of a positive bird or mosquito pool does not by itself constitute evidence of an imminent threat to human health and warrant mosquito adulticiding. Adulticiding should be considered only after consideration of the risk to human health by taking into account multiple factors, including:

- documentation of the presence of mosquito-borne viruses in the area
- the abundance and species of the mosquito populations
- mosquito minimum infection rate (MIR)
- the density and proximity of human populations
- the time of year and weather conditions
- accessibility to the area where the mosquito vector is located
- rapidity of the response required as determined by the seriousness of the public health threat
- the potential impact on people and the environment

Alert Level 4

High Risk of Human Outbreak

Spring, summer, or fall: Surveillance indicates high risk of human infection as shown by indicators such as high dead bird densities, sustained high mosquito infection rates, multiple positive mosquito species, equine or mammal cases indicating escalating epizootic transmission, a human case with high levels of bird, mosquito, or equine infections, areas with early WNV activity that experience epidemic conditions in past years.

Surveillance Response	Education Response	Mosquito Control Response	Communications
Recommended Response for Local Health Jurisdictions and other local entities			
<ul style="list-style-type: none"> Continue surveillance activities described in Alert Level 3. Stop collection of birds and focus on mosquito collection for testing. Continue to monitor and record dead bird sightings and locations. Consider active surveillance for persons hospitalized with mosquito-borne infections. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 3. Expand community-based activities to increase attention to mosquito-borne disease transmission risk. Increase visibility of public messages. Engage key local partners (government officials, religious leaders) to speak about mosquito-borne disease. Maximize visibility of public education through mass media and use of local leaders and organizations to target information for identified high risk groups and areas. 	<ul style="list-style-type: none"> Continue control activities described in Alert Level 3. Continue larval control efforts. Consider implementation of adult mosquito control, targeted at areas of potential human risk if surveillance indicates the risk is likely to increase and vector mosquito species are abundant.² If feasible, monitor the effectiveness of spraying on target mosquito populations if adulticides are used. 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 3.
Recommended Response for Washington State Department of Health			
<ul style="list-style-type: none"> Continue coordination of the statewide WNV monitoring system, as described above. Continue other surveillance activities described in Alert Level 3. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 3. Maximize visibility of public education through mass media and use of local leaders and organizations to target information for identified high risk groups and areas. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue control activities described in Alert Level 3. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 3. DOH ZD Program alerts internal and external partners that one or more LHJs are at high risk of a human outbreak. Establish regular <i>meet me</i> calls (teleconference meetings) with affected LHJs.

Alert Level 5

Outbreak in Progress

Spring, summer, or fall: Multiple confirmed human infections; conditions favoring continued transmission to humans (for example, persistent high infection rate in mosquitoes, and continued avian mortality due to mosquito-borne viruses).

Surveillance Response	Education Response	Mosquito Control Response	Communications
Recommended Response for Local Health Jurisdictions and other local entities			
<ul style="list-style-type: none"> Continue surveillance activities described in Alert Level 4. No bird collection recommended. Continue to monitor and record dead bird sightings and locations. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 4. Enhance risk communication about adult mosquito control. Emphasize urgency of personal protection through community leaders and media. Emphasize use of repellents at visible public events. 	<ul style="list-style-type: none"> Continue control activities described in Alert Level 4. Consider intensifying adult mosquito control program by widespread application of low-volume adulticides and repeat adulticide applications in areas of high risk or human cases.² Monitor efficacy of spraying on target mosquito populations. If outbreak is widespread and covers multiple jurisdictions, consider a coordinated, widespread aerial adulticide application.² 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 4.
Recommended Response for Washington State Department of Health			
<ul style="list-style-type: none"> Continue coordination of the statewide WNV monitoring system, as described above. Continue surveillance activities described in Alert Level 4. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue education activities described in Alert Level 4. Enhance risk communication about adult mosquito control. Emphasize urgency of personal protection through community leaders and media. Emphasize use of repellents at visible public events. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue mosquito control activities described in Alert Level 4. Provide training, technical assistance, consultation, and coordination as needed. 	<ul style="list-style-type: none"> Continue communications activities described in Alert Level 4. DOH ZD Program alerts internal and external partners that one or more LHJs are experiencing an outbreak in progress. Establish frequent <i>meet me</i> calls with affected LHJs.



Appendix A – Mosquito Biology

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Basic Mosquito Information

Basic Mosquito Biology

Mosquitoes are insects belonging to the order Diptera, the True Flies. Like all True Flies, they have two wings, but unlike other flies, their wings have scales and their mouthparts form a long, piercing-sucking proboscis. Only female mosquitoes bite animals or humans for a blood meal to nourish their eggs. Males differ from females by having feathery antennae and mouthparts not suitable for piercing skin. Nectar is the principal food source for males.

There are over 3,000 species of mosquitoes throughout the world, of which 174 species and subspecies occur in North America. Currently, 44 species are known to occur or have been identified in various collections in Washington.

Mosquitoes from six genera are found in Washington and include *Aedes*, *Ochlerotatus*, *Culex*, *Culiseta*, *Anopheles*, and *Coquillettidia*.

The mosquito goes through four distinct stages in its life cycle: Egg, Larva, Pupa, and Adult. Each of these stages is easily recognized by its unique appearance.

The entire life cycle from egg to adult can occur in as little as seven days or a month or longer, depending on the species and time of year. Mosquitoes can also over-winter in all stages except the pupal stage. Hibernating adults seek shelter in basements, culverts, or other protected areas, and larvae can over-winter in permanent pools or containers of water. Eggs deposited in mud or soil along rivers, or in flood plains, will hatch when flooded, even after several years.

Egg

Eggs are laid singly (*Anopheles*, *Coquillettidia*, and *Ochlerotatus*) or in rafts (*Culex* and *Culiseta*) on the surface of the water, or singly on damp soil that is subject to flooding (*Aedes* and *Ochlerotatus*). Most eggs hatch into larvae within 48 hours, however eggs laid on soil do not hatch until flooded and may remain viable for several years.

Larva

Larvae, commonly known as “wigglers,” live in the water and come to the surface to breathe. They shed their skin four times, growing larger after each molting. The stages between molts are called instars. Most larvae have siphon tubes for breathing and hang from the water surface. *Anopheles* larvae lack an air tube and lay parallel to the water surface to obtain oxygen through a breathing opening. Larvae of *Coquillettidia* have a modified respiratory siphon that allows them to attach to plants, such as cattails, below the water surface and get oxygen through their siphon from the stalks or roots of the plant. Larvae constantly feed on microorganisms and organic matter in the water and on the fourth molt change

into pupae. The larval stage generally lasts 7-14 days, depending on water temperature and food resources.

Pupa

Pupae, also called “tumblers,” do not feed. The metamorphosis of the mosquito into an adult is completed within the pupal case. After one to four days, the pupal case splits and the adult mosquito emerges.

Adult

The newly emerged adult rests on the surface of the water for a short time to allow itself to dry and body parts to harden. The wings must be dry before it can fly. The average life span of an adult mosquito is one to two months. They generally have a flight range of .5 to 2 miles, but some species travel up to 20 miles. Both female and male mosquitoes are nectar feeders. However, females must also seek a blood meal for the production of eggs. Mating usually takes place before the female seeks a blood meal, after which she looks for a suitable place to lay her eggs.

Mosquito Life Cycle

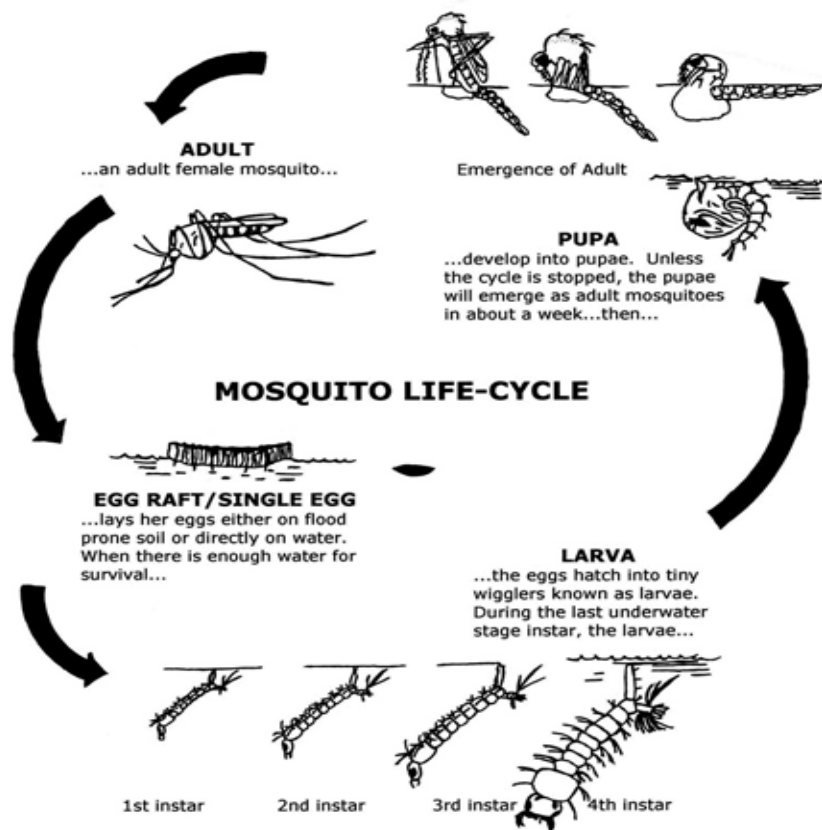



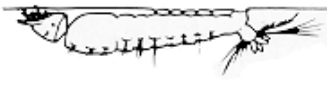
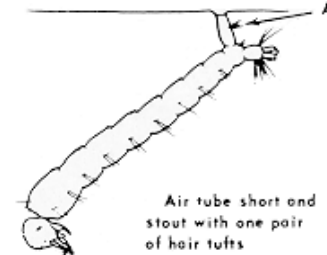
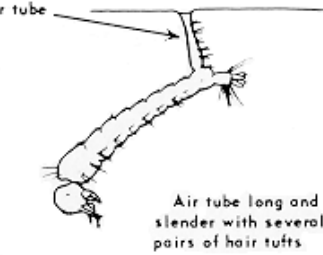


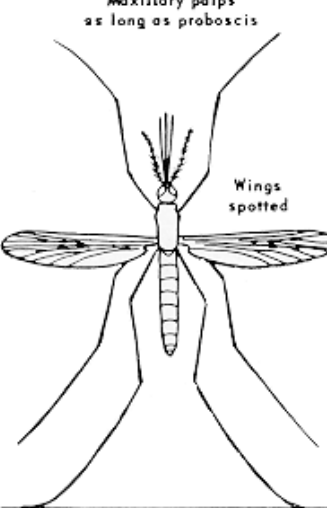
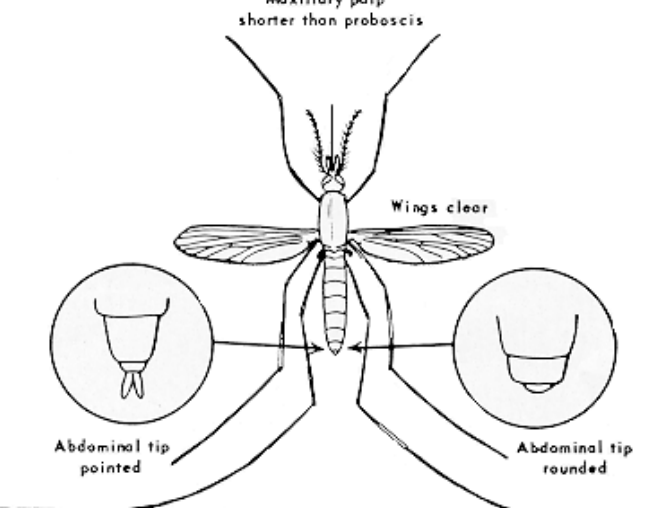
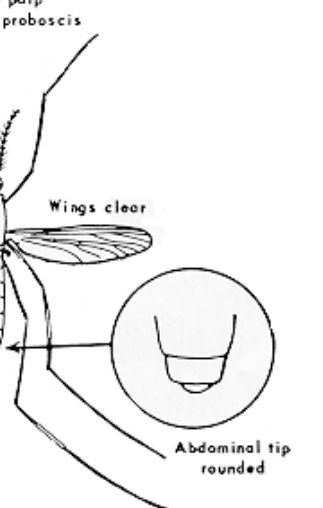





Image Credit: West Umitilla Vector Control District

Principal Characters for Identifying Mosquitoes

	ANOPHELES	AEDES	CULEX
EGGS	 <p>Laid singly Has floats</p>	 <p>Laid singly No floats</p>	 <p>Laid in rafts No floats</p>
LARVAE	 <p>Rest parallel to water surface No air tube Head rotated 180° when feeding</p>	 <p>Rest at an angle Air tube short and stout with one pair of hair tufts</p>	 <p>Rest at an angle Air tube long and slender with several pairs of hair tufts</p>
PUPAE			<p>Pupae differ slightly</p>
ADULTS	 <p>Maxillary palps as long as proboscis Wings spotted</p>	 <p>Maxillary palps shorter than proboscis Wings clear Abdominal tip pointed</p>	 <p>Maxillary palps shorter than proboscis Wings clear Abdominal tip rounded</p>
	 <p>Proboscis and body in one axis</p>		 <p>Proboscis and body in two axes</p>



Mosquito Habitats in Washington

Mosquito problems can occur in variety of habitats. In some areas problems are caused by floodwater, in others by irrigation, and some are related to ponds and artificial containers. Mosquitoes that breed in permanent or semi-permanent pools will usually be found in most localities.

Floodwater

Aedes vexans and *Ochlerotatus sticticus* develop in large numbers along the borders of the Columbia and other rivers, creating one of the most important mosquito problems in this state. The larvae hatch in the spring or early summer when the streams overflow brushy bottomlands and cottonwood swales where the eggs have been laid. The eggs of these species are dormant when temperatures remain below 45°-50° F. Partial dormancy of the eggs may continue until some time in June so that only some of the eggs are hatched by floods occurring in April or May.

In some seasons the larger rivers may rise, recede, and rise again to cover the same egg beds and produce an additional hatch. In other seasons, two or three successive rises may occur, each of which is higher than the last. Females that emerge in the first hatch may lay eggs that will hatch in the second or third rises of the river. Most of the eggs are laid between the ten and 20 foot levels, and some of the eggs that are not flooded during a series of low flood crest years remain viable for as long as 4 years.

Large *Ae. vexans* and *Oc. sticticus* breeding areas have been managed effectively in the past by controlling water levels above the Bonneville Dam. Dikes have prevented flooding in other areas. Clearing of brush has been of value in some locations. However, control of the major section of these floodwater habitats must often be accomplished with insecticides targeting the larvae or adult mosquitoes.

Irrigation Water

Breeding places for several mosquito species are provided by irrigation water. *Ochlerotatus dorsalis*, *Ae. vexans*, *Oc. melanimon*, and *Oc. nigromaculis* are among the most important species that may develop when water is applied and allowed to stand for a week or 10 days. Other species such as *Culex tarsalis*, *Culiseta inornata*, and *Anopheles freeborni* may be produced if water remains for longer periods. Tremendous numbers of mosquitoes breed in many areas where uncontrolled irrigation is practiced. Applications of insecticides to control the larvae or adults that have emerged are effective, but are not substitutes for proper irrigation water management.

Elimination of standing water is effective in preventing mosquito larval development. Application of insecticides may be necessary for larval habitats that cannot be drained.

Tidal Waters

Ochlerotatus dorsalis is the only species that can breed in large numbers in both fresh and salt water in the northwest. The larvae develop in some coastal areas where potholes are filled by the higher tides or where water levels fluctuate in permanent or semi-permanent pools. Leveling, drainage, or similar practices are effective in preventing breeding, but such areas must be properly maintained. Insecticide control of the larvae or adults may be necessary where these methods are inadequate or ineffective. *Ochlerotatus togoi* has also been found in coastal areas, including San Juan, Island, Skagit, and Whatcom counties. This species prefers rock holes just above the high tide line, where *Oc. togoi* larvae have been found in pools of pure seawater along rocky shorelines.

Snow Water

In many high mountain meadows, and also at lower levels, mosquitoes breed in pools caused by snow melt. Development may require several weeks at higher elevations. *Ochlerotatus communis*, *Ae. cinereus*, *Oc. hexodontus*, *Oc. aboriginis*, *Oc. fitchii*, and *Oc. increpitus* are the most common species found in these locations. Usually there is only one generation per year, but the large numbers that may be produced are a severe annoyance to those who are working or seeking recreation in these areas.

Elimination of breeding areas by drainage or maintenance of constant water levels is practical in some situations. Insecticide applications might have to be made by hand or by plane because of inaccessibility to heavy ground equipment.

Ponds and Artificial Containers

The mosquitoes that lay their eggs on the water are usually found where water is present continuously during the season or at least for several days. Such locations include natural permanent ponds, log ponds, semi-permanent ponds of various types, and artificial containers. *Culex tarsalis*, *Cx. pipiens*, *Cx. stigmatasoma*, *An. freeborni*, *An. punctipennis*, *Cs. incidens*, and *Cs. inornata* are commonly found in such places. *Culex tarsalis* and *Cx. pipiens* develop in large numbers in log ponds. *Culex pipiens* also develops in large numbers in sewer drains, catch basins, and water left in artificial containers. *Coquillettidia perturbans* is found in permanent water in swamps and marshes that have emergent or floating vegetation.

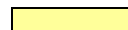
Insecticides are often used effectively to control most of these species, except those breeding in artificial containers. Water standing in barrels, cans, old tires,

and other receptacles should be emptied. Larvae of *Coquillettidia perturbans* are difficult to control because they are attached to the roots of plants. Eliminating host plants, if possible, or using slow release insecticide briquets are methods that have shown to be effective in controlling this species.

Mosquito Species by County, Western Washington

County	<i>Aedes cinereus</i> *	<i>Aedes vexans</i> *	<i>Anopheles earlei</i>	<i>Anopheles freeborni</i> *	<i>Anopheles occidentalis</i>	<i>Anopheles punctipennis</i> *	<i>Coquilleidia perturbans</i> *	<i>Culex apicalis</i>	<i>Culex boharti</i>	<i>Culex pipiens</i> *	<i>Culex stigmatasoma</i> *	<i>Culex tarsalis</i> *	<i>Culex territans</i> *	<i>Culiseta impatiens</i> *	<i>Culiseta incidens</i>	<i>Culiseta inornata</i> *	<i>Culiseta minnesotae</i>	<i>Culiseta morsitans</i> *	<i>Culiseta particeps</i>	<i>Ochlerotatus aboriginis</i>	<i>Ochlerotatus aloponotum</i>	<i>Ochlerotatus campestris</i>	<i>Ochlerotatus canadensis</i> *	<i>Ochlerotatus cataphylla</i>	<i>Ochlerotatus communis</i>	<i>Ochlerotatus dorsalis</i> *	<i>Ochlerotatus excrucians</i>	<i>Ochlerotatus fitchii</i> *	<i>Ochlerotatus flavescens</i>	<i>Ochlerotatus hexodontus</i>	<i>Ochlerotatus impiger</i>	<i>Ochlerotatus implicatus</i>	<i>Ochlerotatus increpitus</i>	<i>Ochlerotatus intrudens</i>	<i>Ochlerotatus japonicus japonicus</i> *	<i>Ochlerotatus melanimon</i> *	<i>Ochlerotatus nevadensis</i>	<i>Ochlerotatus nigromaculis</i> *	<i>Ochlerotatus pullatus</i>	<i>Ochlerotatus sierrensis</i>	<i>Ochlerotatus spencerii idahoensis</i>	<i>Ochlerotatus sticticus</i> *	<i>Ochlerotatus togoi</i>	<i>Ochlerotatus ventrovittis</i>	
Western Washington																																													
Clallam	X	X	X	X	2		X	X	X	X	X	X	X	X	X	X				X				X	X	X												2		X	X	2			
Clark	X	X	X	X	X		X	X	X	X	X	2	X	X		2									2																	X	X	X	
Cowlitz	X	X	X	X	X	2	2	X	X	X	X	X	X	X	X	4	2	4	4	X			4	X	5							X	X									2	X	X	
Grays Harbor	X			X	3	X		X	X	X	X	4	X		5	3	3	X																							5	4			
Island	X	3	X	X	2			X	X	X	2	X	X	X	3	2	2	X	2						1	X					X					3				3		X	2		
Jefferson	X	X		4	3			X	X	X	X	X	X	5				X	5					X	X	X	X	X			X									X	X	5			
King	X	4	1	X	X	X	4	X	X	X	1	X	X	X	5	2	X	X					X	X	X	X		X						2	1					X	X	X			
Kitsap		3		X	2			X		X	3	X	X	3		X	4	X	X					X	X	X		X			X										X				
Lewis	X			X				X	X	X	X	X	X	X					X				X		X		X	X			X										X				
Mason	X	X			3			X	X	X									X	X				X	X	X														X					
Pacific	X	X			X			X	X	X	2	X		3	3	2	X						X	X	X						X										X	X	X		
Pierce	X	X	X	X	X			X	X	4	X	X	X	X	X	4	3	X	X	X			X	X	X	X	X	X	X	X	X	X	X	X	5		X	X	X	X	X	X	X	X	
San Juan				3	3			3	3	3	3	X	3										X	X																	X		X		
Skagit	X			3				X	X	X	X	X							X	X				2	X	X		X	X	X	X	X								X	X	X	X	X	
Skamania	X	X	X	X	3		3	X	X	3	X	X	X						X				X	X	X	X	X	X	X	X	X	X								X	X	X			
Snohomish	X	X	4	5	3	X	2	X		X	X	X	X	X	4	X	3	X							4	X	X														3			2	X
Thurston	X			X	X			X	X	X		X	1		1	3	X	X							X	X	X														X		3		
Wahkiakum			5	5	5			5		X	5	5		5	5										5																5				
Whatcom	X	X	X	X	X			4	X	X	X	X	X	X	3	3			X	X				X	X	X	X															3	X	2	

*WNV-positive mosquito species based on national surveillance reported to CDC, 1999-2005:



Last Revised 06/20/07

New findings for: 1 - 2001 2 - 2002 3 - 2003 4 - 2004 5 - 2005

Previous findings: X

The matrix shows the known distribution of mosquito species by county for western Washington through the year 2005. Previous findings are based on mosquito surveillance conducted by Washington State Department of Health in the 1960s and 1970s. In 2007, previous findings were updated to reflect historical findings presented in the *Distribution of Mosquitoes in Washington State*, 2007 William J. Sames et al., Journal of the American Mosquito Control Association (unpublished, as of June 2007). New mosquito species findings, which had not been identified during earlier surveillance efforts, are presented by the surveillance year 2001 through 2005, when the species were first detected.

Not included in matrix: Eight species reported for Washington are based upon one or two county records and in some cases one or few specimens. The species, *Culex erythrothorax*, *Culex restuans*, *Culex salinarius*, *Ochlerotatus pionips*, *Ochlerotatus provocans*, *Ochlerotatus punctor*, *Ochlerotatus trivittatus*, and *Othropodomyia signifera*, are not included in the matrix.

Mosquito Species by County, Eastern Washington

County	<i>Aedes cinereus</i> *	<i>Aedes vexans</i> *	<i>Anopheles earlei</i>	<i>Anopheles freeborni</i> *	<i>Anopheles occidentalis</i>	<i>Anopheles punctipennis</i> *	<i>Coquillettidia perturbans</i> *	<i>Culex apicalis</i>	<i>Culex boharti</i>	<i>Culex pipiens</i> *	<i>Culex stigmatasoma</i> *	<i>Culex tarsalis</i> *	<i>Culex territans</i> *	<i>Culiseta impatiens</i> *	<i>Culiseta incidens</i>	<i>Culiseta inornata</i> *	<i>Culiseta minnesotae</i>	<i>Culiseta morsitans</i> *	<i>Culiseta parviceps</i>	<i>Ochlerotatus aboriginis</i>	<i>Ochlerotatus aloponotum</i>	<i>Ochlerotatus campestris</i>	<i>Ochlerotatus canadensis</i> *	<i>Ochlerotatus cataphylla</i>	<i>Ochlerotatus communis</i>	<i>Ochlerotatus dorsalis</i> *	<i>Ochlerotatus excrucians</i>	<i>Ochlerotatus fitchii</i> *	<i>Ochlerotatus flavescens</i>	<i>Ochlerotatus hexodontus</i>	<i>Ochlerotatus impiger</i>	<i>Ochlerotatus implicatus</i>	<i>Ochlerotatus increpitus</i>	<i>Ochlerotatus intrudens</i>	<i>Ochlerotatus japonicus japonicus</i> *	<i>Ochlerotatus melanimon</i> *	<i>Ochlerotatus nevadensis</i>	<i>Ochlerotatus nigromaculis</i> *	<i>Ochlerotatus pullatus</i>	<i>Ochlerotatus sierrensis</i>	<i>Ochlerotatus spencerii idahoensis</i>	<i>Ochlerotatus sticticus</i> *	<i>Ochlerotatus togoi</i>	<i>Ochlerotatus ventrovittis</i>
Eastern Washington																																												
Adams		4		4			4			X	X					X										X										4	X							
Asotin				X							X	X																																
Benton		X		X	X	X	4			X	X	X			X	X	X				X		5	X	X	X	X						X			X	X	X	X	X	X	X		
Chelan	X	X		X		X				X	X	X	X	X	X	X	X	1		X				X	X	X	X	X	X	X	X	X	X	X				X	X	X	X	X	X	X
Columbia		X		X							X										X													X										
Douglas		X									X		X												X											X								
Ferry	X	X		X			X			2	X	X		2					X				X	X		X	X	X	X	X	X	X	X									2		
Franklin		4		3	X	4	4			X	X										5				4							1			X	X	3		X					
Garfield						X					X			X																														
Grant	X	1		X		X				X	X	X	X	X	X									X	X	X	X	X	X				X				X	X	2					
Kittitas	X	X		X	X	X				X	X	X	X	X	X	X			X					X	X	X	X	X				X					X	X		X	X			
Klickitat		X								3	X	X		3	3				X														3						3					
Lincoln		X		X				X	3	X			X	3										X	X	X	X						X						3					
Okanogan	X	X	X	X	X	X	3			X	X	X		X	X							X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Pend Oreille	X	X	X	X	X					X	X	X					X						X	X		X	X	X	X	X	X	X	X	X	X					X	X	X	X	
Spokane	X	X	3	X		X	2			X	X		1	X	5	X			X	X	X	X	X	X	X	X	X	X				X			3			X	4		X			
Stevens	X	X	X	X	X					X	X	X	X	X									X	X	X	X	X					X				X	X		3					
Walla Walla	X	X		X		X	3			X	X	X	1	1	X	X				X															2		X	3		2				
Whitman		X		X	X	X				X	X		X	X					3		X				3							X							X		4			
Yakima	X	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	

*WNV-positive mosquito species based on national surveillance reported to CDC, 1999-2005:

Last Revised 06/20/07

New findings for: 1 - 2001 2 - 2002 3 - 2003 4 - 2004 5 - 2005

Previous findings: X

The matrix shows the known distribution of mosquito species by county for western Washington through the year 2005. Previous findings are based on mosquito surveillance conducted by Washington State Department of Health in the 1960s and 1970s. In 2007, previous findings were updated to reflect historical findings presented in the *Distribution of Mosquitoes in Washington State*, 2007 William J. Sames et al., Journal of the American Mosquito Control Association (unpublished, as of June 2007). New mosquito species findings, which had not been identified during earlier surveillance efforts, are presented by the surveillance year 2001 through 2005, when the species were first detected.

Not included in matrix: Eight species reported for Washington are based upon one or two county records and in some cases one or few specimens. The species, *Culex erythrorhax*, *Culex restuans*, *Culex salinarius*, *Ochlerotatus pionips*, *Ochlerotatus provocans*, *Ochlerotatus punctor*, *Ochlerotatus trivittatus*, and *Othropodomyia signifera*, are not included in the matrix.

In 2001, mosquito larvae collected from "Lucky Bamboo" shipments in Benton County, were reared and identified by CHPPM-West as *Ochlerotatus j. japonicus*. CDC confirmed the identification in 2002. No further collections have been made in Benton County, therefore this one time finding is not included in the matrix.



Potential Amplifying and Bridge Vector Mosquitoes for West Nile Virus in Washington

The following mosquitoes, present in Washington, are species from which West Nile virus (WNV) has been isolated and/or West Nile viral ribonucleic acid (RNA) has been detected in parts of the country where WNV is present. Isolation of WNV or detection of West Nile viral RNA in a mosquito species does not necessarily incriminate that species as a competent vector of the disease. It is only an indication that the species has come into contact with the West Nile transmission cycle. Some mosquitoes may play a part in amplifying the virus in the environment, while others may help amplify the virus and/or be competent bridge vectors capable of transmitting disease to animals other than birds. Based on information from other parts of the country, *Cx. pipiens* and *Cx. tarsalis* will be our state's most efficient WNV bridge vector species.

Aedes cinereus

This mosquito can be found in a wide range of larval habitats but is most frequently found in woodland and open meadow pools and cattail swamps. In some mountain areas it is the predominant species. It does not travel far from its larval habitat. It is known to rest in the grass or underbrush. This species is a serious pest and will bite any time during the day. It is known as an ankle biter because it focuses on the lower extremities. It may be a late hatcher in the colder climates.

Aedes vexans

One of the most common floodwater mosquitoes, this species is found in large numbers in irrigated and inland floodwater areas. Eggs are laid in mud and hatch when flooded in the spring or early summer. Several hatches may occur each season if water levels recede and rise. However, the eggs will remain viable for several years if flooding does not occur. This species has a flight range of over 20 miles. *Ae. vexans* are vicious biters of humans and domestic mammals. They feed during the day, but more commonly bite at dusk and dawn. They are most abundant along the bottom lands of the Columbia River and other rivers.

Anopheles freeborni

Also called the Western malaria mosquito, this mosquito is abundant in irrigation waters, roadside ditches and other sunlit pools and along the margins of creeks and rivers from spring till frost. The females overwinter in sheltered areas and emerge in midwinter to lay eggs. They may have several generations per year.

An. freeborni feed on humans and large mammals from dusk to dawn. They have a flight range of about ten miles.

Anopheles punctipennis

The larvae of this species are usually found in pools of fresh water containing vegetation that are around for several weeks such as natural ponds or log ponds. They may also be found in cool grassy pools along creeks and rivers and in artificial containers and other environments associated with *Cx. tarsalis* and *Cx. pipiens*. They are aggressive day and dusk biters and feed on large mammals, including cows and horses, as well as humans. They do not fly far from their breeding sites.

Coquillettidia perturbans

This species breeds in marshes, ponds and lakes that have a thick growth of cattails or other aquatic vegetation. Larvae attach to the stalks of vegetation and do not need to rise to the surface to breathe, making control difficult. They have been found in storm water ponds. They bite birds, mammals, and humans. This species enters houses, are fierce, painful biters and are active primarily in the evening, but will bite during the day in shady places. They generally over-winter as larvae.

Culex pipiens

This mosquito, commonly called the northern house mosquito, was probably introduced to the west coast of North America during the 1800s. It is widespread in Washington and readily enters houses on warm summer nights. Although they occur in rural environments, they reach their greatest numbers in urban and suburban areas. Their flight range is about one mile. They breed in catch basins, storm water ponds, clean and polluted ground pools, ditches, animal waste lagoons, log ponds, and other waters rich in organic matter. This species also deposits eggs in artificial containers that hold water, such as tin cans, tires, and birdbaths. Larvae may be present from spring through fall. *Culex pipiens* primarily feeds on birds, but will also feed on mammals, including humans and dogs. This mosquito will play a significant role in the amplification of WNV and can serve as a competent bridge vector.

Culex stigmatasoma

Larvae are occasionally found in clean water, but primarily in polluted water on farms, sewage plants, and artificial containers. Females rarely feed on humans.

Culex tarsalis

This mosquito is probably the most widespread species in Washington, found in nearly all counties. The larvae develop in many types of permanent and semi-permanent waters such as log ponds, storm water ponds, ditches, marshes and pools in grasslands and woodlands in either clean or polluted waters. Larvae may also be found in artificial containers such as tires, tin cans, and ornamental ponds. This species prefers to feed on humans, domestic animals and birds. It has a reported flight range of over ten miles. Primarily an evening and early morning biter, this species is the most important vector of western equine encephalitis and St. Louis encephalitis. Mated females may hibernate over winter and emerge in early spring. The wide distribution and opportunistic feeding behavior of *Cx. tarsalis* make it a strong candidate for serving as an amplifying and bridge vector of WNV in Washington.

Culex territans

These mosquitoes are found in clear unpolluted waters. The females lay their eggs on the bank and rely upon rain to flush the eggs onto the water surface. This mosquito generally feeds on amphibians and other small animals inhabiting their resting areas. They rarely attack humans. Adults have the ability to withstand cold winter temperatures and have several generations per year.

Culiseta impatiens

This species favors woodland and timbered habitats. It is found along logging roads, ditches and other small pools. This mosquito is an early season biter. The females overwinter with one generation per year.

Culiseta inornata

This species is widely distributed and primarily breeds in woodland pools, but can also be found in any slow moving or stagnant water. However, it is seldom found in artificial containers. It can be found at elevations up to 6,000 feet in very cold water. *Culiseta inornata* is a serious pest of livestock due to its long breeding season and wide distribution in irrigated areas. It is often associated with *An. freeborni* and *Cx. tarsalis* and is a competent vector of western equine encephalitis.

Culiseta morsitans

These mosquitoes inhabit semi-permanent woodland swamps and bogs containing grasses and cattails. They are suspected of over-wintering in the egg and larval stages and occasionally as adults. Birds are their preferred hosts. Larvae are produced in early spring.

Ochlerotatus canadensis

Found in woodland pools filled by melting snow or rain, this species is one of the first to emerge in the spring from overwintering eggs. It can be a serious pest in shaded areas near its breeding site and the adults live for several months. It feeds on a broad range of animals including large and small mammals, birds and reptiles.

Ochlerotatus dorsalis

The Pale Marsh Mosquito is named after its whitish-grey appearance. Larvae are found in brackish and saline waters as it breeds along the edges of bays, marshes, lakes, and flooded pastures. Females prefer to feed on large mammals like cattle and horses, but readily bite humans. They are vicious biters throughout the day, with major activity occurring towards dusk. They are so aggressive and persistent that livestock tend to move away from areas where they are numerous. A strong flyer, *Oc. dorsalis* often disperses 20 miles or more from its breeding sources.

Ochlerotatus fitchii

This mosquito is widely distributed in Washington and readily bites humans. Larvae are found in large, grassy pools and pools created by spring snow melts. It has also been reported from cranberry bogs. Eggs overwinter and there may be two generations a year.

Ochlerotatus japonicus

A newly discovered species in Washington, initially found in 2001 in King County, *Oc. japonicus* is expanding its range, having been identified in Pierce and Snohomish counties in 2003. Cowlitz County found *Oc. japonicus* in adult mosquito traps in 2006. Larvae are found primarily in artificial containers, depressions filled with water, tires, birdbaths and coastal rock pools. This species is a daytime biter making avoidance difficult. The distribution of this species will be better understood as more surveillance is undertaken. Habitat reduction by eliminating standing water and water in containers is important in controlling the population of this species.

Ochlerotatus melanimon

Larvae are found in irrigated pastures, fields and saline pools. They are similar to and may be associated with *Oc. dorsalis*. Adults will bite humans as well as domestic animals. Their flying range has been reported to be ten miles.

Ochlerotatus nigromaculis

This species is often associated with *Oc. dorsalis* and other species found in irrigated or flooded meadows. This species is a major pest for large mammals and humans. It is a strong flyer with a 20-mile range.

Ochlerotatus sticticus

This species is serious pest to humans and domestic mammals in Washington. It breeds in large numbers along the brushy flood plains of the Columbia River and other similar locations. Eggs are viable for several years on inactive flood plains. Adults may disperse up to 20 miles.



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Appendix B – Mosquito Surveillance

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Establishing a Mosquito Surveillance Program

Effective mosquito control begins with a sustained, consistent surveillance program that targets pest and vector species, identifies and maps their immature habitats by season, and documents the need for control. Records should be kept on the species composition of mosquito populations prior to enacting control of any kind and to allow programs to determine the effectiveness of control operations.

Surveillance is a critical component of an integrated pest management (IPM) and control program. Knowing when and where to treat mosquito populations reduces the use of pesticides and allows for targeted application when pesticides are applied, thereby reducing waste as well. All components of the integrated management program must be monitored for efficacy using best practices and standard indices of effectiveness.

A comprehensive mosquito surveillance program must include larval and adult sampling components, a mapping/record keeping component, and a data analysis component. To provide useful data, the surveillance program must be sustained and maintain a consistent effort over several seasons. There are several advantages of mosquito-based surveillance, including:

- May provide the earliest evidence of disease activity or transmission in an area.
- Helps establish information on potential mosquito vector species.
- Provides an estimate of vector species abundance.
- Provides baseline data that can be used to guide emergency control operations.
- Allows evaluation of control methods.
- Gives quantifiable information on virus infection rates in different mosquito species.
- Provides quantifiable information on potential risk to humans and animals.

Surveys are essential for the planning, operation and evaluation of an effective mosquito-control program, whether for the prevention of mosquito-borne diseases or to reduce mosquito populations to levels permitting normal activities without undue discomfort. Initial surveys identify the species of mosquitoes present and provide general information on locations, densities and disease potential. With this knowledge it may be possible to determine life cycles and

feeding preferences; predict larval habitats, adult resting places and flight ranges; and perhaps even make preliminary recommendations for control programs.

A formal surveillance program is one in which routine monitoring of mosquito presence is conducted, thus providing long term data of mosquito activity. Surveillance activities address adult and larval population density and species composition, rainfall and tide monitoring, and breeding site locations. Such surveillance does not determine the absolute population of mosquitoes, but it can show fluctuations in relative mosquito abundance and diversity over time in the various habitats visited.

Larval surveillance involves sampling a wide range of aquatic habitats for the presence of pest and vector species during their developmental stages. Trained inspectors should collect larval specimens on a regular basis from known larval habitats and perform systematic surveillance for new sources. Properly trained mosquito identification specialists can separate nuisance and vector mosquito species. Responsible control programs target vector and nuisance populations for control and avoid managing habitats that support benign species.

Adult mosquito surveillance is used to monitor species presence and relative abundance of adult mosquitoes in an area. Information derived from adult mosquito surveillance programs using standardized and consistent surveillance efforts provide information essential to monitoring potential vector activity, setting action thresholds, and evaluating control efforts. Adult surveillance can provide information on pathogens such as West Nile, St. Louis, and western equine encephalitis viruses, which may be circulating within a mosquito population.

The detection and measurement of pest mosquito populations has often proved easier by routine light trap and human bait collections of adults than by larval sampling. However, treating larval habitats has, in general, remained a more effective method of reducing mosquito populations than the application of adulticide sprays. Larval surveys have consequently continued to be important in assessing population size and the impact of control measures.

Additional specialized surveillance may be conducted to detect arboviral presence in birds and mosquito populations. This information not only provides justification for habitat reduction and insecticide applications, but it also serves as an ongoing indicator of the effectiveness of these activities and continually adds to the database of knowledge concerning mosquitoes in the area.

Mapping

Reasonably accurate and comprehensive maps are essential in conducting a mosquito control operation. Maps provide information for field survey and control activities, program evaluation, and reporting and budgeting purposes.

They show elevations, streets, roads and railroads, as well as ponds, lakes, streams, sewage lagoons, flooded woodlots and other breeding areas. They are used for orientation and for locating and plotting larval breeding places and adult sampling stations. When large areas are involved, a master map may be needed for planning drainage and other field operations. The master map will indicate the treatment areas, the possible flight range of mosquitoes from breeding sites, and the potential degree of penetration into populated areas. Larval and adult sampling stations can be indicated by symbols and numbers. Counts made at these stations at weekly or biweekly intervals provide information for current evaluation of the mosquito problem at any time by indicating the abundance of mosquitoes, species involved, flight range and habitat, and disease potential. This information identifies areas requiring high priority for treatment.

Narrative descriptions, sometimes necessary for exact location description, are simplified whenever possible. For example, “N.W. corner of Axil Street and Dipper Road” is a brief description that leaves no doubt as to the location. There may be some areas that are difficult to accurately locate (such as marshlands). However, maps can be subdivided into numbered or named areas for easy reference, and Global Positioning System (GPS) coordinates are very reliable. Some common methods of subdividing maps involve the use of geographical features, artificial grids or a combination of both to set boundaries on areas that are indexed for easy reference and filing. To avoid cluttering, the larger areas may be further subdivided by the use of transparent overlays, again employing geographical features or a grid. Once the area of inspection is delineated by reference to index numbers, additional location data can be conveyed clearly by the use of cards that include a rough sketch of the area or incorporated into a Geographic Information System (GIS) format.

Record Keeping

In order to avoid comparing dissimilar parameters, inspections should be consistent both in method and location. Keeping clear, accurate records is as important as the data gathering itself. Surveillance records are managed in a manner that ensures subsequent inspections can be conducted in a similar manner by others less familiar with the area. They usually include the inspector's name, date of inspection and exact location in addition to the data collected. Data-recording forms and devices promote uniformity, which makes records easier to read, interpret and summarize, and serve as a reminder to the inspector to record all pertinent information. In the absence of data recorders, standardized formats lead to more consistently accurate transcription of the data into the permanent records. Washington DOH mosquito surveillance data forms can be found online at <http://www.doh.wa.gov/ehp/ts/Zoo/WNV/LocalHealth.html>.



Adult Mosquito Collection Methods

The objectives of adult sampling are to determine species composition, population age structure, what species are currently active, where the greatest numbers are located and, if conducting arboviral or pathogen surveillance, what species are carrying what pathogens. Adult surveys also can provide data on seasonal and spatial distribution of the vector(s). Depending on the type of information desired, different collection methods and equipment may be required.

Resting Populations

Sampling adult resting populations provides a better cross-section of the overall population. Resting adults comprise both fed and unfed individuals at all stages of their adult life.

Adults of many mosquito species are inactive during the day, resting quietly in dark, cool, humid places. An index of the population density can be obtained by carefully counting the number of adults found in a resting station. These sampling sites are also a source of specimens for mosquito-borne virus tests. Sampling resting adults usually provides a representative sample of the population: collections include teneral and post-teneral unfed, blood-fed, and gravid females, as well as males. Population age structure also is more representative. However, different species and different gonotrophic stages may prefer different types of resting sites. Sampling resting populations is usually time consuming, especially when looking for natural resting sites. The number of specimens collected per unit of effort may be low compared to other collection methods.

Resting mosquitoes are most easily collected with an aspirator, but can be collected with a sweep net or drop nets, which are useful for collecting specimens resting in grass or low vegetation.

“Natural” Resting Sites: Natural resting sites include any location not specifically constructed to serve as shelter for mosquitoes. Examples are storm sewers and culverts, bridges, houses, porches, barns, stables, chicken houses, privies, rodent burrows, tree holes and vegetation. With experience, the suitability of shelters as adult mosquito resting stations is easily evaluated. Collections must be standardized for accurate comparison of results.

“Artificial” Resting Sites: Artificial resting stations may be constructed when suitable natural resting stations are not available. Many different types of artificial shelters have been used, including red boxes (wooden boxes painted black on the outside and red on the inside), red cloth shelters, and privy-type shelters. These shelters should be placed in shaded, humid locations near

suspected breeding places or in other known congregation sites. Most species enter such shelters around dawn in response to environmental factors such as changes in light intensity and humidity, and ordinarily do not leave until dusk. Artificial shelter boxes, one cubic foot in size with one side open and painted red on the inside, have been used successfully for several species in the United States. Walk-in red boxes have also been shown to be very effective for capture of *Culex tarsalis* and other species.

Active Populations

Sampling active mosquito populations provides a picture of what species are currently seeking a host. Sampling of active mosquitoes can also provide information on the location of activity as well as when during the day or night various species are most actively seeking a blood meal.

Baits and Attractants: Animal-baited and CO₂-baited traps disproportionately attract host-seeking females. This is the segment of the population of greatest interest for mosquito-borne virus surveillance. The type of bait used is important in trap performance. Often there is significant variability in attractiveness depending on the bait used, and this may affect trap performance. Other considerations are the duration of collection and time of day (especially important for species with a narrow host-seeking window). A final consideration is the decision whether to let mosquitoes feed or not (important if specimens are to be used for blood meal identification). Specimens can be removed from the trap periodically with a hand-held aspirator.

CO₂-baited traps rely on the dissipation of dry ice (or bottled CO₂) as the attractant, imitating CO₂ release by the host in animal-baited traps. Another material, 1-octen-3-ol (octenol), has been used, either alone or with CO₂, as an attractant in bait traps. Octenol is a derivative of gases produced in the rumen of cows and has been shown to be an effective attractant to some species (such as *Oc. japonicus*).

Drop Nets and Tent Traps: These traps normally are left open or are suspended above the animal being used as bait. After a set period, the openings are closed or the net lowered, and the trapped mosquitoes are collected. Trap size can vary, depending upon the size of the bait being used. Mosquito bed nets can be modified for use as a tent trap or drop net.

Small Animal Bait Traps: Some animals are more attractive than others to certain mosquitoes. For example, light traps baited with birds can be a more attractive than if baited with CO₂ or a mammal, while other species are less attracted to bird baits and more attracted to a CO₂-baited trap.

Dry Ice and Hand Aspirator: Some mosquito species can be collected by having the collector stand over or near a small block of dry ice. Females that are

attracted by the CO₂ can be collected with a net or hand-held aspirator as they fly around the collector's legs.

Landing/Biting Counts: Landing/biting collections, usually using humans or horses, sample selected portions of the mosquito population, particularly in studies to incriminate specific vectors or in other research applications. When using human bait, consideration must be given to the potential health risks involved. During epidemics, it is advisable to restrict these activities to naturally immune or immunized individuals, or refrain from conducting landing/biting counts until the epidemic is over.

Light Traps

There are two primary functions that light traps provide in mosquito surveillance programs. One function is to provide a historical record of mosquito abundance and species presence in an area. Historical data show fluctuations on a year to year basis as well as fluctuations over the span of one season. This type of information can be used to document the impact of mosquito control activities and provide the justification for additional control efforts in an area. Light trap records are especially useful for program budgeting and acquiring water management and pesticide use permits.

The second function of light traps is to provide rapid information on mosquito abundance and species composition for planning and directing day-to-day mosquito control activities.

Light trap data are used:

- to determine or to help the need, the timing, and/or the location of pesticide applications and to monitor the results of those pesticide applications.
- to help determine the cause of repeated mosquito complaints in a given area.
- as a supplement or backup to more expedient surveillance techniques such as landing or bite counts.

Additionally, because males typically emerge 24 hours before females emerge, finding males in the sampling trap is a good indication of female emergence within 24 to 48 hours.

Many mosquito species are attracted to light, making it possible to sample adult populations between dusk and dawn. There is, however, considerable variation in the relative attractiveness of different mosquito species to light. Some species are not attracted to light at all, and may even be repelled (such as *Cx. quinquefasciatus*). The catch is also influenced by many factors, including light source, wavelength and intensity. Generally, light traps do not reflect the

abundance or presence of species that are not attracted to light or are only active during the day. In addition, mosquito species that inhabit wooded areas are less attracted to light traps than those which prefer open areas. Because of these differences, light trap collections should be used in conjunction with other population sampling methods. Competing light sources (including moonlight, roadside lights, and “urban glow”), fan size and speed, and presence or absence of screens also affect trap performance. Another factor to be aware of is that light traps only sample the flying population.

Trap placement (height, location in relation to trees and other cover, proximity to breeding sites), can greatly influence the composition of species collected. Some trial and error placement is generally necessary in locating good trap placement sites. Traps are typically hung 5-6 feet off the ground, which is a satisfactory height for the majority of species being collected. Mosquitoes that do not seek hosts between dusk and dawn will either be missed or under-represented. Adults that feed during the day can be trapped by simply adjusting the trap hours of operation to include a representative portion of daylight collection time.

Whenever possible, light traps should be placed in the same location year after year for general surveillance practices. For greater accuracy in measuring population changes, newly installed traps should be located where mosquito populations are high. When installing the trap, be mindful of exterior lighting such as spot lights, windows and exhaust vents. The trap should be placed in an open area, 30 feet or more away from buildings, but close to trees and shrubs. Attempts should also be made to protect the trap from prevailing winds. It should not be placed near other lights, in areas subject to strong winds, or near industrial plants emitting smoke or fumes. Traps should be operated on a regular schedule from one to seven nights per week, from just before dark until after daylight.

Light trap captures are improved dramatically with the addition of dry ice as a carbon dioxide attractant. The amount of dry ice as well as the type of container used to hold it will affect the amount of carbon dioxide released over time. In most instances, a five pound block of dry ice is sufficient to cover the normal dusk to dawn trapping period. Insulated containers are available from the manufacturer or home-made containers can be easily constructed from cloth or paint cans or plastic buckets insulated with flexible foam padding and with holes poked into the bottom edges to allow the CO₂ to dissipate. Dry ice containers should be hung in a position to allow the carbon dioxide to release directly next to or slightly above the trap. When host-seeking mosquitoes enter the stream of gas, they are drawn into the trap by the fan.

In regions where dry ice is difficult to obtain, CO₂ canisters, sweaty socks, or a caged animal, bird or mammal, can be used as an attractant.

New Jersey Light Trap: The New Jersey-type light trap was developed in the early 1940's. It was widely used in adult surveys because of its attraction to mosquitoes and its durability and was a standard device used by mosquito control agencies in the United States.

At present, this trap continues to be used, but its design places certain restrictions on its use. Conventional usage requires that an electric outlet be available to power the trap. Additionally, because many locations where electrical power is available are places where competing light sources exist, the effectiveness of the trap is reduced. Added to this is the fact that the higher wattage bulb in New Jersey traps tends to attract a lot of "trash" insects, which cause considerable damage to mosquito specimens once they are caught. As a result, the trap is no longer used as extensively as it once was and has been replaced by portable light traps as a preferred method of surveillance.

CDC/EVS Light Traps: The Centers for Disease Control (CDC) miniature and encephalitis vector survey (EVS) light traps were developed for greater portability. These traps can be taken to remote areas that could not otherwise be sampled by a trap dependent upon electricity. They are commonly operated with four 1½-volt "D" cell flashlight batteries, or one 6-volt motorcycle battery, which provides sufficient power for one night's trapping. They are light and easily disassembled for transport. Both traps are fitted with a nylon collecting bag (or a cardboard carton). In this way, the catch is captured and held alive until the specimens can be collected for processing.

Because portable light traps were designed to use an extremely small light bulb, their light output is much weaker than the 25 watt bulb used in the New Jersey light traps. Neither the CDC or EVS light trap competes well with other light sources; smaller catches may result during a full moon. Mosquitoes can be collected with light as the only attractant, but the addition of dry ice greatly enhances the trap's capabilities with increases of 400 – 500% in overall catch reported. Dry ice also increases the number of species captured by 20 – 25% and improves the ratio of blood-fed and parous individuals for mosquito-borne virus surveillance. Traps can also be used with CO₂ and no light, and be effective at collecting species like *Cx. tarsalis* without attracting many of the other insects that are normally attracted by the light. See Guidelines for Using Encephalitis Vector Survey Light Traps (page 69).

Propane Traps: A recent development in mosquito traps has been a variety of propane powered traps. Propane traps, baited with octenol and/or CO₂, have been shown to be very attractive to some mosquito species. However, attraction varies between species, and even by the type of propane trap used, resulting in a biased sample. Therefore, while propane traps can provide large catch rates, they should be used in conjunction with other trap types to obtain a more comprehensive survey of the mosquito species in the area.

The control of adult mosquitoes begins with proper surveillance. A dry ice-baited, portable light trap is an efficient, reliable surveillance tool and can be used to assess a variety of issues, like a homeowner's complaint, the success of an adulticide, or to gather viral activity information.

Oviposition Traps

Oviposition traps sample populations of mosquito females looking for a suitable location to lay their eggs, also called gravid females. Since the gravid population has fed at least one time, these individuals are more likely to be infected. This reduces the work involved in processing mosquito pools for virus isolation. Minimum infection rates (MIRs) will, on average, be higher than those obtained from light trap catches. Traps can be separated on the basis of whether or not they retain the ovipositing (egg-laying) females or allow them to escape.

Ovitrap: Ovitrap only sample eggs, but the number of *Culex* rafts can be used to estimate the ovipositing (and therefore recently-fed) adult female population. Several trap designs are available for various mosquito genera and species. In general, ovitraps for *Aedes* species are small, while traps for *Culex* species are larger, and usually have an attractant or infusion.

Reiter Gravid Trap: The Reiter gravid trap samples female *Culex* mosquitoes as they come to lay their eggs. It is selective for females that have already taken at least one blood meal. If mosquitoes are being collected for virus isolation, there is a higher probability of collecting infected mosquitoes. Gravid trap counts might also have a higher correlation with disease transmission. See Guidelines for Using Gravid Traps (page 71).



Larval Mosquito Collection Methods

The main objective of mosquito larvae sampling is to detect larval habitats and assess any significant changes in larval density resulting from control measures. Different species of mosquitoes prefer different habitats in which to lay their eggs. Mosquito larvae and pupae are found in a great variety of different habitats, ranging from large expanses of water such as swamps and flooded meadows to small collections of water, like those found in plant axils, snail shells, fallen leaves, and tires. Numerous types of mosquito habitats exist in Washington. These include areas prone to flooding, irrigation sites, tidal water ponds, snowmelt pools, ponds, marshes, artificial containers, tree holes, storm water retention ponds, and catch basins.

Larval surveillance is an essential component of an effective mosquito surveillance and control program and larval surveys have many important functions. They are used to determine the locations and seasons that mosquitoes use specific aquatic habitats and, when specimens are identified and counted, the information can be used to determine species composition and population densities. The information can be used to determine optimal times for application of larval control measures, including chemicals, biologicals, draining or impounding. It can also be used to help forecast the need for adult mosquito control and to help assess the effectiveness of both chemical and biological control measures.

Routine larval surveillance data can be useful in interpreting adult mosquito surveillance data. For example, if larval surveys indicate 95-100% control by larvicides and yet the number of adults does not decline, one can suspect, in the absence of reinfestation, that an important larval concentration was missed.

Larval surveillance should be conducted at each known site one to two times per week during the mosquito season. However inspectors should always be on the lookout for potential new larval habitats. Data should include site identification, weather at time of collection, the number of larvae collected per dip, how many dips were taken, the average number of larvae per dip, and what species were collected. Larvae can be identified in their larval form, stored in vials of alcohol for later identification, or reared to adults for identification.

Sampling Methods

Dipping: The most commonly used larval collection method is the "standard dipper," usually a plastic, white pint to quart-sized scoop-on-a-handle. The number of dips made, the number of larvae collected per dip, and the numbers of larvae found, by instar if warranted, are recorded and representative sample specimens are transferred by pipette into small vials of alcohol for later identification. With most species, it is possible to get a rough idea of the breeding

activity by computing the average number of larvae of each species per dip. The number of dips required will depend on the size of the area and the relative larval density, but for convenience is often in multiples of ten. Inspections should be made at weekly or biweekly intervals during the mosquito breeding season, as areas that are entirely negative at one time may rapidly become heavily infested.

The species of mosquitoes being looked for and the type of habitat being sampled will, in part, determine the sampling method used. Thus, it is important that field personnel know the preferred breeding habitats and seasonal occurrence of species known or suspected to be present within an area.

When surveying for mosquito larvae, approach the habitat cautiously to avoid disturbing larvae at the water's surface. Vibrations from heavy footsteps, casting a shadow or moving vegetation that contacts the water may be enough to cause larvae to dive to the bottom. Try to approach the water while facing the sun, quietly and slowly, moving vegetation only as necessary.

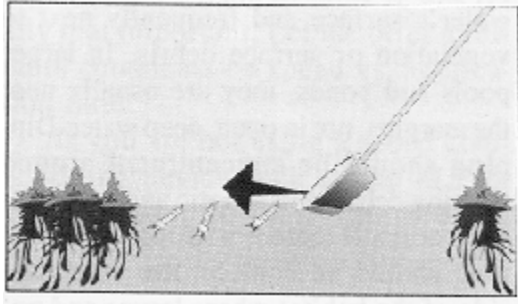
Mosquito larvae of most genera, particularly *Culex*, *Aedes*, *Ochlerotatus*, and *Anopheles*, are usually found at the water's surface and frequently next to vegetation or surface debris. In larger ponds, they are usually near the margins, not in open, deep water. Dipping should be concentrated around floating debris and aquatic and emergent vegetation. If there is a strong wind, dipping should be done on the windward side of the habitat where larvae and pupae will be most heavily concentrated. Look for larvae and pupae before beginning to dip, if possible. If it is raining on the water's surface, wait until the rain stops.

Each water body may contain a number of different microhabitats which could contain different mosquito species. Microhabitats are such places as under tree roots, within clumps of emergent vegetation, under floating or overhanging vegetation and in open water. Learn to recognize different microhabitats within an area and sample as many as possible in order to obtain an accurate picture of the area's species composition.

There are seven basic methods to dip for mosquito larvae. Which method or methods you use depend on the genus or genera of mosquitoes you suspect may be present and on the habitat, microhabitat and weather conditions.

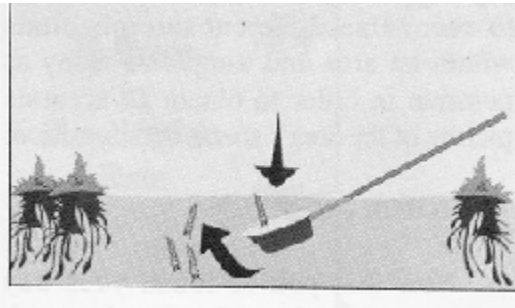
1. **Shallow Skim.** The shallow skim consists of submerging the leading edge of the dipper, tipped about 45 degrees, about an inch below the surface of the water and quickly, but gently, moving the dipper along a straight line in open water or in water with small floating debris. End the stroke just before the dipper is filled to prevent overflowing. The shallow skim is particularly effective for *Anopheles* larvae that tend to remain at the surface longer than *Aedes* and *Culex*. *Anopheles* are usually associated with floating vegetation and debris.

Figure 1. Shallow Skim



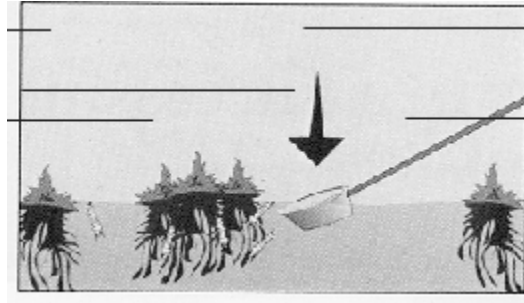
2. **Complete Submersion.** Used in open water, with or without, floating objects. Many mosquito larvae, particularly those of the genera *Aedes* and *Ochlerotatus*, are very active and usually dive below the surface quickly if disturbed. In this case, a quick plunge of the dipper below the surface of the water is required, bringing the dipper back up through the diving larvae. Bring the dipper up carefully to avoid losing the larvae in the overflow current.

Figure 2. Complete Submersion



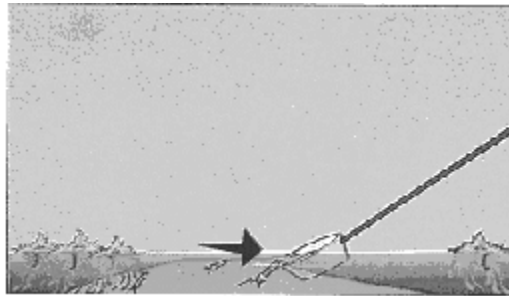
3. **Partial Submersion.** Use for sampling at the edges of emergent vegetation. Push the dipper, tilted at about 45 degrees, straight down adjacent to the vegetation. This causes the water around the vegetation to flow into the dipper, carrying the larvae with the flow. There is no need to move the dipper horizontally. Pull the dipper up before it is full.

Figure 3. Partial Submersion



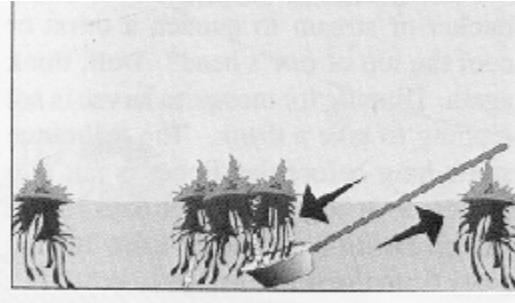
4. **Flow-In.** Use in very shallow water. Larvae can be collected by pushing the dipper into the substrate of the pool and letting the shallow surface water, debris and larvae flow into the dipper. Do not move the dipper horizontally.

Figure 4. Flow-In



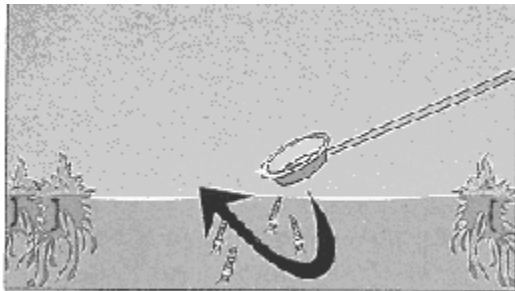
5. **Scraping.** Use for sampling larvae that may be under floating or emergent vegetation. This method is used in habitats that contain clumps of vegetation such as tussocks of sedges, floating mats of cattails or water lettuce or other plants that are too large to get in the dipper, or clumps of submerged vegetation such as hydrilla or bladderwort. Dip from the water in towards the vegetation and end by using the dipper to scrape up against the base or underside of the vegetation to dislodge larvae. This method is usually more effective if the bottom of the dipper is screened and it is often used to sample for *Coquilletidia* mosquitoes.

Figure 5. Scraping



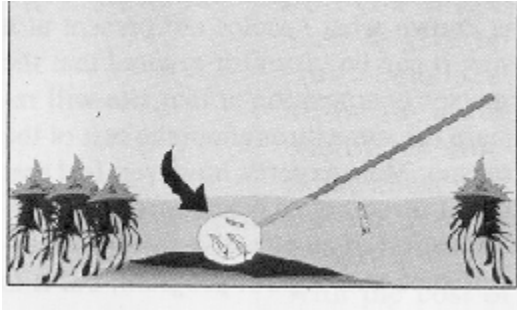
6. **Simple Scoop.** It consists of simply scooping a dipperful of water. This is probably the most commonly used method, particularly by new inspectors, and it is often the method referred to in much of the literature as "the standard dipping procedure." While it can be successfully used to collect *Culex* larvae, it is still not the method of choice.

Figure 6. Simple Scoop



7. **Background.** This technique is especially useful in woodland pools and other shallow water or when larvae are disturbed and dive to the bottom. Submerge the dipper completely to the bottom litter and slowly move it around. The darker mosquito larvae and pupae will stand out against the background of a white or aluminum dipper. Once larvae appear in the dipper, just lift it upward.

Figure 7. Background



One or more of these methods, properly used, can determine the mosquito species composition of most aquatic habitats, excluding those whose openings are smaller than the dipper. Turkey basters or other syringe-type suction devices, allow access to difficult places like plant axils, treeholes, tree root holes, rock pools, tires, and a variety of artificial containers used by some mosquito species. Other tools, such as a vial, measuring spoon, or tea strainer can be used in these habitats utilizing the same sampling techniques described above.

Submerged Vegetation: *Coquillettidia perturbans* larvae have a modified siphon, toothed and spur-like, which is inserted into submerged roots or stems of plants to obtain air from the plant itself. *Coquillettidia* larval habitats are usually in freshwater swamps, grassy pools or along lake margins. Favorite plants include cattails and other emergent vegetation.

Because these larvae are rarely found at the water surface, they can not be sampled by the usual dipper technique. Sampling can be done by uprooting the plants and immediately placing them in a bucket of clear water. Vigorously shake the submerged parts to dislodge any larvae attached to the root mats and plant stems. After mud and debris have settled, larvae and pupae can be collected either from the water surface or can be pipetted from the bottom mud.



Guidelines for Using Encephalitis Vector Survey (EVS) Light Traps

The following guidelines are presented to improve the reliability of light trap usage in county mosquito surveillance programs and encourage uniformity of light trap data throughout the state.

Light Trap Operation

- Consider two traps as the minimum number in most situations and compare your data to detect differences that may have been due to outside influences.
- Operate the light traps on a regular schedule, between 2 and 7 nights per week from dusk to dawn, depending on mosquito populations and risk of mosquito-borne diseases.
- Trap at least one hour prior to dusk until one hour after dawn to insure that surveillance is conducted during the primary host-seeking periods for most species.
- Use a clear bulb in the trap to make your data comparable with that of other mosquito control agencies.
- Whenever possible, use traps in combination with a dry ice supplement. A 4-5 lb. block in an insulated container will mimic a large mammal's respiration and last long enough to cover the usual dusk to dawn trapping period.
- Hang the dry ice adjacent to the trap to draw mosquitoes as close as possible to the collection fan.
- Begin trapping at the beginning of mosquito season each year and continue the trapping program through the month of September or later, depending on mosquito activity. Washington mosquito activity usually begins in April and winds down in October; however, some species may carry activity into November while others hibernate as adults in secluded refuges.

Light Trap Placement

- Be mindful of exterior lighting when installing a light trap and try to place the trap in open areas, away from buildings at a height of 5-6 feet above the ground unless specific information is needed on canopy dwellers. For most nuisance species, this height will provide a reliable indication of activity.

- Try to set the traps along the edges of habitats to increase trapping efficiency. A trap located strictly in one ecosystem or habitat may exclude certain species. Trapping along the edge of a swamp, for example, will provide a picture of those species found not only in the swamp, but also in the nearby upland.

Other Considerations

- Be aware that the males in the collection provide useful information. A large number of male mosquitoes indicate that a brood of females is about to emerge in that area.
- Be aware that there is considerable variation in the relative attractiveness of different mosquitoes to light and evaluate your light trap data accordingly. Other species may be present that need to be sampled using a different method.
- Be aware that differences do exist in the host seeking behavior of some species and that modifications of these general guidelines may be necessary to get complete surveillance data. Species that feed only during the daytime will not be accurately represented in dusk-dawn collections. A species that hunts in tree canopies will not be accurately sampled by a trap that is suspended five feet from the ground. Whenever possible, become familiar with the host seeking habits of the mosquitoes being surveyed. Don't be afraid to experiment!
- Whenever possible, identify the collections within 24 hours.



Guidelines for Using Gravid Traps

Trap Placement

- Locate the trap in or near residential areas in order to collect container-breeding *Culex* species. Traps should be located in areas protected from extreme environmental conditions (for example, wind and direct sun) and in secure areas (not conspicuous) where they will not be disturbed or vandalized.
- Appropriate trap sites include: utility yards, window and stair wells, storm drains, boatyards, animal stables, transformer pits, cluttered backyards, tire storage yards, and cemeteries.
- It is desirable to have some type of overhead cover (shrubs or refuse) so that the tub is not easily flooded in the event of rain. Punch holes in the sides of the container so excess water drains out.
- Locate traps where they can be visited daily. If after several visits the trap does not appear productive, move it to an alternate location. Remember the primary goal is to collect blood-fed female mosquitoes. It is less of a priority to maintain consistent sites that may have poor yields.

Trap Set Up

- If using the trap for the first time, be sure to rid the plastic tubs of potential insect repellent properties associated with the chemical components found in some plastics. This can be done by immersing the tubs in a muddy pond for several days or filling with water and leaving it out in the open for a week or two.
- At least two days before trapping, mix, in a gallon jug or jerry can, approximately one cup of rabbit pellet food (available from pet or feed stores) per one gallon of aged (allowed sit out for two to three days to permit the chlorine to evaporate) water. Let the concentrate incubate in a protected (inaccessible to mosquitoes) location. Change the media out as often as necessary to ensure a fresh mixture for each night of trapping.
- At the trapping site put approximately $\frac{1}{4}$ gallon of the premixed rabbit food concentrate to the tub and add aged water collected from a nearby natural source (such as a pond or stream) to bring the water level up to within two inches of the bottom edge of the fan housing tube.
- Position the trap bracket securely over the center of the tub and slide the collection bag (or screened tub) over the top of the trap tube. Be sure the

bag is not askew and that it remains properly positioned, even if a breeze picks up.

- Attach the battery to the terminal wires and make sure it is securely positioned, and test the trap making sure the fan turns freely and draws the air from below.
- Place the trap out in the late afternoon (for *Culex* spp). Gravid traps can also attract some daytime species (for example, *Ochlerotatus* spp., *Aedes* spp.) if placed out earlier in the day.
- Assign the trap a number and note its location on a map.
- Depending on your situation, it may be advisable to label the traps with a warning stating “West Nile Virus Surveillance – Do Not Touch” and include your organization’s name and a contact phone number.

Trap Servicing and Specimen Collection

- Visit the trap early the following day. Carefully remove the trap bag (or screened tub) containing mosquitoes and replace with an empty one. Tie off the open end, and if the bag is not easily hung in the servicing vehicle, place net props (such as a tongue depressors) around the bag so that it does not collapse and the mosquitoes are not crushed.
- Note in a field notebook or on your data sheet the general number of mosquitoes taken from each particular trap (to be verified later in the laboratory) and any other relevant information.
- The water can be used for multiple trap nights within a week (top it off with aged water to make up for evaporation), but dump the water at the end of one week of trapping and make a new batch of the rabbit pellet food water as described above. If this is not done, eggs potentially laid could develop thereby contributing to mosquito breeding in the vicinity. (Note: prior to emptying water, the water surface can be examined for the presence of eggs and, if present, collected in specimen jars for rearing and species confirmation).
- It is best to trap at the beginning of the week; Monday, Tuesday, and Wednesday. This will allow time for sorting and overnight mailing specimens before the weekend. This also allows for adjustments if there is some reason you can’t trap on a given night (storms or holidays). Remember, the attractant concentrate needs to incubate for at least two days, so it should be prepared no later than Friday for the following week.



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Appendix C – Bird & Mammal Surveillance

Dead Bird Surveillance for West Nile Virus [75](#)

Equine West Nile Virus [77](#)

Human West Nile Virus [79](#)



Dead Bird Surveillance for West Nile Virus

Dead birds testing positive for WNV are a sensitive indicator of viral activity in a geographical area and can provide an early warning to local communities.

Various approaches for dead bird surveillance have been used by states where WNV outbreaks have occurred. These activities have ranged from reporting and testing only dead crows in some states to testing a broader range of bird species in other states.

Based on national dead bird surveillance data since 1999, testing of Corvidae species (jays, magpies, ravens, and crows) and raptor species (hawks, owls, eagles, and other birds of prey) will increase the probability of detecting WNV in Washington. Corvids and raptors are very susceptible to the virus and have a high mortality rate. Therefore, submission of these birds for WNV testing is a top priority.

Birds testing positive for WNV have been most commonly found dead. The small percentage of sick birds observed showed symptoms such as weakness, lethargy, tremors, inability to walk, fly, perch, or hold their wings normally against their body, and were easily approachable by humans. These signs of generalized illness are not unique to WNV infection. Raptors and corvids infected with WNV and who appear sick, are often in fair to poor physical condition as well, with loss of fat and muscle mass particularly noticeable in the flight muscles (breast).

Reports of a substantial number of dead birds, which are not corvids or raptors, in the same location are typically shown to have died from causes other than WNV. Such die-offs are frequently associated with pesticide or natural poisoning incidents. Birds associated with these die-offs are not considered suitable specimens for WNV surveillance. We recommend reporting bird die-offs to the Washington State Department of Fish and Wildlife or United States Fish and Wildlife Service office in your area.

We also expect that reports of dead birds may be associated with concerns about avian influenza. Guidance on how to handle dead bird calls and concerns about avian influenza is available. Please visit the following Web site for more information: <http://www.doh.wa.gov/ehp/ts/Zoo/WNV/deadbirdsAI.pdf>.

Birds submitted for WNV testing, should be either a corvid or raptor and dead less than 48 hours, with the carcass intact. If the carcass has an odor, is soft and mushy, has skin discoloration, feathers or skin that easily rubs off, or has maggots present, it is too decomposed for testing. Birds which meet requirements for testing may be frozen for storage and shipping. View the collecting and

shipping protocol for WNV dead bird monitoring at <http://www.doh.wa.gov/ehp/ts/Zoo/WNV/DBSurProt.pdf>. The dead bird reporting form is available at <http://www.doh.wa.gov/ehp/ts/Zoo/WNV/birdreportform.pdf>.

In addition to testing selected specimens, it is also necessary to record reports of dead birds that are not tested. Significant correlations have been made between areas with high densities of reports and areas where human cases appear one to four weeks later. Studies in several states have shown that, in urban and suburban areas, as many as 75% of human cases occurred near locations where clusters of dead corvids (primarily crows) were found. Use the same form submitted with laboratory specimens to record information about dead birds sighted and reported, but not tested. Retain and map these reports to evaluate areas of heavier sighting.



Equine West Nile Virus

Horses can also be important sentinels of WNV epizootic activity and human risk, at least in some geographic regions. In addition, equine health is an important economic issue. Therefore, surveillance for equine WNV disease should be conducted in jurisdictions where horses are present. Veterinarians, veterinary service organizations, and state agriculture departments are essential partners in any surveillance activities involving equine WNV disease.

Surveillance

The goals of equine disease surveillance are to use data on horse WNV disease cases to assess the threat of human disease, identify geographic areas of high risk, and assess the need for and timing of interventions.

An advantage of equine disease surveillance is that horses are highly conspicuous, numerous, and widely distributed in some areas. Using ill horses as an early indication of WNV has been advantageous in some areas. However, many areas have had positive horse cases just prior to or simultaneously with human WNV disease cases. Another disadvantage of equine disease surveillance is that widespread use of horse WNV vaccines may decrease the incidence of WNV disease and therefore the usefulness of horses as sentinels.

Reporting of Possible Cases

The Washington State Department of Health (DOH), Washington State Department of Agriculture (WSDA), and the United State Department of Agriculture (USDA) are requesting that veterinarians report equine encephalitis cases of unknown etiology for possible mosquito-borne disease testing.

To report possible West Nile virus in equines, veterinarians and local health jurisdictions should contact the:

Washington State Department of Agriculture
State Veterinarian's Office
360.902.1878

United States Department of Agriculture Office
360.753.9430

In support of local WNV diagnostic efforts, the Washington Animal Disease Diagnostic Laboratory (WADDL) provides diagnostic testing for antibodies in equine serum and cerebro-spinal fluid (CSF), viral nucleic acids using polymerase chain reaction (PCR), and antigens using immunohistochemical (IHC) analysis. Practitioners with questions about the diagnosis of suspected equine WNV can contact WADDL at 509.335.9696. Equine cases of suspect WNV will also be tested for rabies at the DOH Public Health Laboratory.

Horse West Nile Virus Vaccine Information

Yearly (annual) vaccination of horses in Washington State against WNV is strongly recommended. If horses are being vaccinated for the first time, they need a series of two doses, three to six weeks apart. If the horse was previously vaccinated with the series, it needs an annual booster before the beginning of the mosquito season. There are several vaccines available.

Horse owners should contact their veterinarians for vaccination and other equine health information.



Human West Nile Virus

West Nile Virus Disease is a Notifiable Condition

West Nile virus infection was first reported in the Western Hemisphere in 1999 during an outbreak in New York City and, since then, has spread east to west across the United States. During 1999 through 2007, over 27,400 human WNV infections resulting in 1065 deaths have been reported to local and state health departments. Over 4,600 cases have been reported in Canada between 2002 and 2007, with over half of those cases occurring in 2007, largely due to an outbreak in the prairie provinces of Saskatchewan, Manitoba and Alberta. Washington had its first in-state acquired infections in 2006.

West Nile virus is usually transmitted to humans by the bite of an infected mosquito. The virus also has been transmitted by blood transfusion, organ transplantation, intrauterine infection, and possibly breastfeeding. Blood banks have been screening blood products for WNV infection since June 2003.

Arboviral (arthropod-borne viral) diseases, including WNV infections, must be reported to local health jurisdictions in Washington State. For more information go to <http://www.doh.wa.gov/Notify/nc/wnv.htm>.

Guidelines for clinicians in West Nile virus disease case reporting to public health can be found at: <http://www.doh.wa.gov/Notify/guidelines/pdf/wnv.pdf>.



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Arboviral Encephalitides

Arthropod-borne viruses, or arboviruses, are viruses that are maintained in nature through biological transmission between susceptible vertebrate hosts by blood feeding arthropods (mosquitoes, psychodids, ceratopogonids, and ticks). Vertebrate infection occurs when the infected arthropod takes a blood meal. The term “arbovirus” has no taxonomic significance. Arboviruses that cause human encephalitis are members of three virus families: the *Togaviridae* (genus *Alphavirus*), *Flaviviridae*, and *Bunyaviridae*.

All arboviral encephalitides are zoonotic, being maintained in complex life cycles involving a nonhuman primary vertebrate host and a primary arthropod vector. These cycles usually remain undetected until humans encroach on a natural focus, or the virus escapes this focus via a secondary vector or vertebrate host as the result of some ecologic change. Humans and domestic animals can develop clinical illness but usually are “dead-end” hosts, because they do not produce significant viremia, and do not contribute to the transmission cycle. Many arboviruses that cause encephalitis have a variety of different vertebrate hosts and some are transmitted by more than one vector. Maintenance of the viruses in nature may be facilitated by vertical transmission (for example, the virus is transmitted from the female through the eggs to the offspring).

Arboviral encephalitides have a global distribution, but there are five main encephalitis viruses in the United States: eastern equine encephalitis (EEE), western equine encephalitis (WEE), St. Louis encephalitis (SLE), La Crosse (LAC) encephalitis, and West Nile virus (WNV), all of which are transmitted by mosquitoes. The three most prominent mosquito-borne diseases in Washington are WEE, SLE, and WNV. Most cases of arboviral encephalitis occur from June through September, when mosquitoes are most active. In warmer parts of the country, where mosquitoes are active late into the year, cases can occur into the winter months.

The majority of human infections are asymptomatic or may result in a nonspecific, flu-like illness. Onset may be gradual or sudden, with fever, headache, myalgias, malaise and occasionally prostration. Infection may, however, lead to encephalitis (and/or meningitis with WNV), with a fatal outcome or permanent neurologic sequelae. Fortunately, only a small proportion of infected persons progress to serious neurological illness.

Because the arboviral encephalitides are viral diseases, antibiotics are not effective for treatment and no effective antiviral drugs have yet been discovered. Treatment is supportive, attempting to deal with problems such as swelling of the brain, loss of the automatic breathing activity of the brain and other treatable complications like bacterial pneumonia. There are no commercially available

human vaccines for these U.S. arboviral diseases. Arboviral encephalitis can be prevented in two major ways: personal protective measures and public health measures to reduce the population of infected mosquitoes. Personal measures include reducing time outdoors, particularly in early evening hours, wearing long pants and long sleeved shirts, and applying mosquito repellent to exposed skin areas. Public health measures include education, surveillance, and the support of integrated pest management practices, which include using insecticides to kill juvenile (larvae) and adult mosquitoes.

Selection of mosquito control methods depends on what needs to be achieved; but, in most emergency situations, the preferred method to achieve maximum results over a wide area is aerial spraying. In many states aerial spraying may be available in certain locations as a means to control nuisance mosquitoes. Such resources can be redirected to areas of virus activity. When aerial spraying is not routinely used, such services are usually contracted for a given time period.

Western Equine Encephalitis

The alphavirus western equine encephalitis was first isolated in California in 1930 from the brain of a horse with encephalitis, and remains an important cause of encephalitis in horses and humans in North America, mainly in western parts of the USA and Canada. In the western United States, the enzootic cycle of WEE involves passerine birds, in which the infection is inapparent, and culicine mosquitoes, principally *Culex tarsalis*, a species that is associated with irrigated agriculture and stream drainages. The virus has also been isolated from a variety of mammal species. Other important mosquito vector species include *Ochlerotatus melanimon* in California, *Oc. dorsalis* in Utah and New Mexico, and *Oc. campestris* in New Mexico. Western equine encephalitis virus was isolated from field-collected larvae of *Oc. dorsalis*, providing evidence that vertical transmission may play an important role in the maintenance cycle of an alphavirus.

Expansion of irrigated agriculture in the North Platte River Valley created habitats and conditions favorable for increases in populations of granivorous birds such as the house sparrow, *Passer domesticus*, and mosquitoes such as *Cx. tarsalis*, *Oc. dorsalis* and *Oc. melanimon*. All of these species may play a role in WEE virus transmission in irrigated areas. In addition to *Cx. tarsalis*, *Oc. dorsalis* and *Oc. melanimon*, WEE virus also has been isolated occasionally from some other mosquito species present in the area.

Human WEE cases are usually first seen in June or July in North America. Most WEE infections are asymptomatic or present as mild, nonspecific illness. Patients with clinically apparent illness usually have a sudden onset with fever, headache, nausea, vomiting, anorexia and malaise, followed by altered mental status, weakness and signs of meningeal irritation. Children, especially those under one

year old, are affected more severely than adults and may be left with permanent sequelae, which is seen in 5 to 30% of young patients. The mortality rate is about three percent.

Washington State's first recorded outbreak of mosquito-borne disease occurred during the summers of 1939-1942. Mosquitoes bred abundantly in many places in central and eastern Washington, especially in areas with careless irrigation practices. In 1942, one of the most significant mosquito surveys was conducted in the Yakima Valley region. During July and August a total of 24,751 mosquitoes were collected, identified to species, and tested for virus content. Of these, 9,466 were *Cx. tarsalis*. A total of 45 viruses were isolated from this species, 41 WEE and four SLE strains. The infection rate demonstrated at least one in 210 mosquitoes collected carried WEE or SLE. The role of *Cx. tarsalis* as a natural vector of these viruses in the Yakima Valley area was thus amply confirmed.

According to published articles and Washington State data, around 200 human cases of mosquito-borne encephalitis (the majority were believed to be WEE) occurred during the 1939-1942 Yakima Valley region outbreaks. During this time period, severe horse epidemics, which affected hundreds (perhaps thousands) of horses, were noted prior to human infections. In the decades that followed the initial outbreaks in the 1930s and 40s, over 100 horses have been confirmed with WEE, and nearly 300 have been suspected cases, including 92 suspected and 23 confirmed horse cases in central and eastern Washington in 1969. After the 1940s, human cases of WEE were sporadic, ranging from 0-3 cases per year. From 1967-1974, 16 people were confirmed with WEE infection. The last confirmed human case of WEE in Washington was in 1988.

St. Louis Encephalitis

In the United States, the leading cause of epidemic flaviviral encephalitis is SLE. St. Louis Encephalitis is the most common mosquito-transmitted human pathogen in the U.S. While periodic SLE epidemics have occurred only in the Midwest and southeast, SLE virus is distributed throughout the lower 48 states. Since 1964, there have been 4,437 confirmed cases of SLE with an average of 193 cases per year (range 4 - 1,967). However, less than 1% of SLE viral infections are clinically apparent and the vast majority of infections remain undiagnosed. Illness ranges in severity from a simple febrile headache to meningoencephalitis, with an overall case-fatality ratio of 5-15%. The disease is generally milder in children than in adults, but in those children who do have disease, there is a high rate of encephalitis. The elderly are at highest risk for severe disease and death. During the summer season, SLE virus is maintained in a mosquito-bird-mosquito cycle, with periodic amplification by peridomestic birds and *Culex* mosquitoes. In Florida, the principal vector is *Cx. nigripalpus*;

in the Midwest, *Cx. pipiens pipiens* and *Cx. p. quinquefasciatus*; and in the western United States, *Cx. tarsalis* and members of the *Cx. pipiens* complex.

Compared to WEE, SLE has had less of an impact on Washington residents, although it continues to persist, at a minimal level, in the natural environment. In 2005, a sentinel chicken maintained by the mosquito control district in Benton County tested positive for SLE. The last two confirmed human cases of SLE in Washington occurred in 1971 and 1972.

West Nile Virus

West Nile Virus is a flavivirus that is, taxonomically, part of the Japanese encephalitis serocomplex, which also includes the SLE, Kunjin and Murray Valley encephalitis viruses, as well as others. West Nile Virus was first isolated in the West Nile Province of Uganda in 1937. The first recorded epidemics occurred in Israel during 1951-1954 and in 1957. Epidemics have been reported in the Rhone delta of France in 1962, Romania in 1996 and in South Africa in 1974.

An outbreak of arboviral encephalitis in New York City and neighboring counties in New York state, in late August and September 1999, was initially attributed to the SLE virus based on positive serologic findings in CSF and serum samples using a virus-specific IgM-capture enzyme-linked immunosorbent assay (ELISA). The outbreak was subsequently confirmed as caused by WNV based on the identification of virus in human, avian, and mosquito samples.

It is not known when and how WNV was introduced into North America. Possible causes include international travel of infected persons to New York, importation of infected birds or mosquitoes, or migration of infected birds. West Nile virus can infect a wide range of vertebrates; in humans it usually produces either asymptomatic infection or a flu-like illness, but can cause severe and fatal infection in a small percentage of patients. People over 50 years of age and the immune compromised are at higher risk of developing the more serious neuroinvasive form of the disease. Other mammals, particularly horses and tree squirrels, are known to develop serious illness due to WNV.

Within its normal geographic distribution of Africa, the Middle East, western Asia, and Europe, WNV has not been documented to cause epizootics in birds. Crows and other birds with antibodies to WNV are common, suggesting that asymptomatic or mild infection usually occurs among birds in those regions. Similarly, substantial bird virulence of SLE virus has not been reported. Therefore, an epizootic producing high mortality in crows and other bird species is unusual for either WNV or SLE virus. For both viruses, migratory birds may play an important role in the natural transmission cycles and spread. Like SLE virus, WNV is transmitted principally by *Culex* species mosquitoes, but also has

been found in *Aedes*, *Anopheles*, *Coquillettidia*, *Culiseta*, *Ochlerotatus*, and other species. In the U.S., the top three most frequent mosquito species that tested positive for WNV in 2006 were *Cx. tarsalis*, *Cx. pipiens*, and *Cx. pipiens* mixed with *Cx. restuans*.

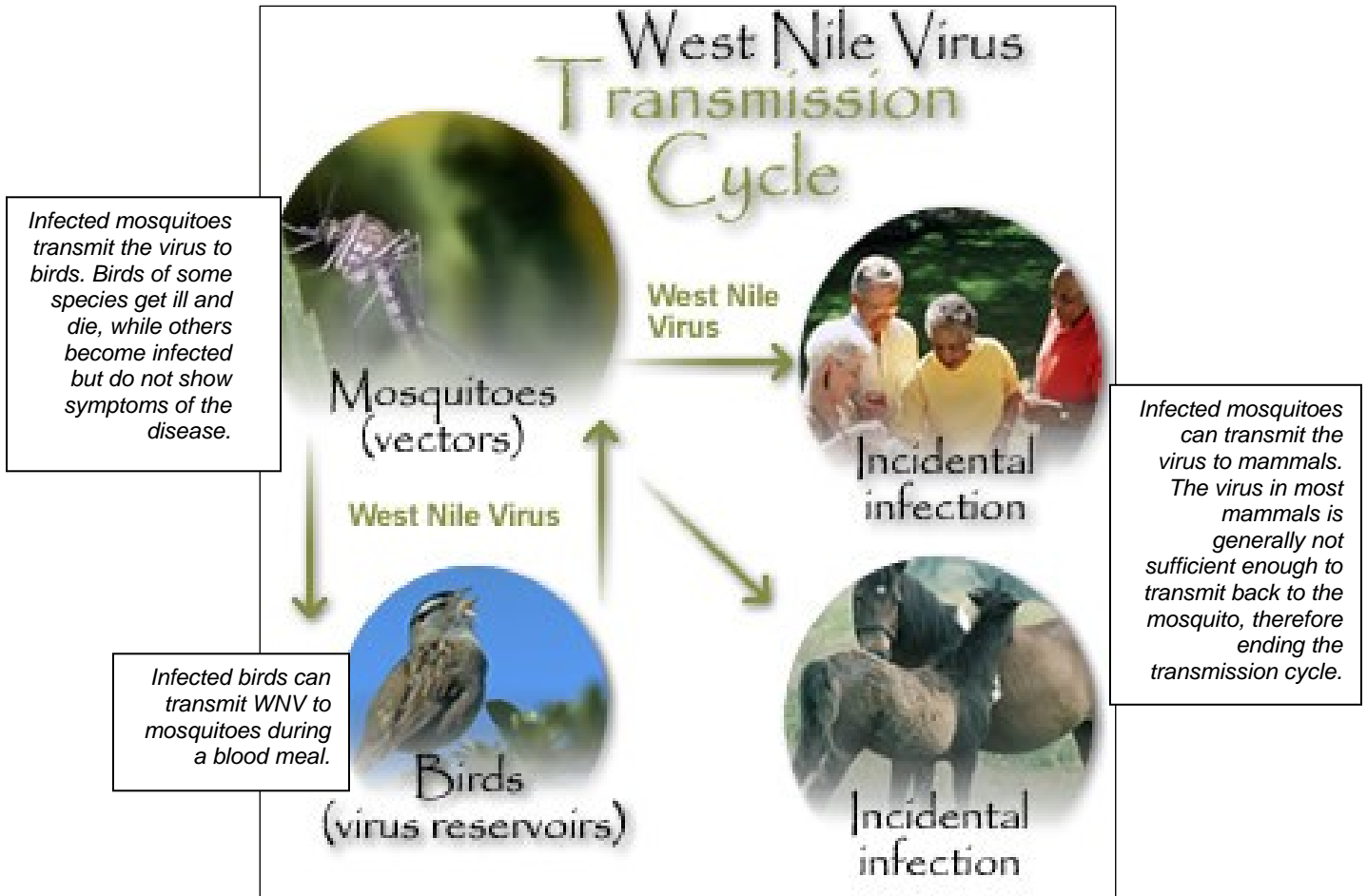
Since its introduction to the U.S. WNV has spread across North America. Surveillance efforts focused on identifying and documenting WNV infection in birds, mosquitoes, and equines as sentinels that could alert health officials to the occurrence of human disease. By the end of 2002, WNV activity had been identified in 44 states. The 2002 WNV epidemic and epizootic resulted in reports of 4,156 human cases of WNV disease (284 deaths), 16,741 dead birds, 6,604 infected mosquito pools, and 14,571 horse cases. Washington State recorded its first WNV activity in 2002 with 4 birds and 2 horses testing positive (page 87). Curiously, Washington did not detect the virus in 2003 or 2004 – why the virus did not establish itself after 2002 was likely due to various environmental factors. However, WNV continued to establish itself elsewhere in the US, and in 2003, 9,862 human cases (264 deaths) were reported, with significant outbreaks occurring in the Great Plain and Rocky Mountain states. By 2004, the majority of the 2,539, human cases occurred in the Southwest. In 2005, there were 3,000 human case reports in the U.S. as the virus established itself in northern California and parts of Idaho and Oregon. Two mosquito pools, one horse, and one bird tested positive for WNV in Washington in 2005. In 2006, there were over 4,200 human cases (over 160 deaths) in the US, with Idaho leading the nation with nearly 1,000 human cases. Washington detected WNV in 3 people, 13 birds, and 6 horses in 2006. In 2007, there were over 3,500 cases of WNV in the U.S., with 109 fatalities. Only 8 horses, 1 dog, and 1 bird tested positive for WNV in Washington in 2007.

Other Arboviral Encephalitides

Many other arboviral encephalitides occur throughout the world. Most of these diseases are problems only for those individuals traveling to countries where the viruses are endemic.

West Nile Virus Transmission Cycle

West Nile Virus Transmission Cycle



Public Health Agency of Canada

WNV Activity in Washington

	2001	2002	2003	2004	2005	2006	2007
Dead Birds Tested	28	325	906	553	660	501	497
WNV-Positive Birds	0	4	0	0	1	13	1
Sentinel Chickens Tested	94	387	435	392	576	441	0
WNV-Positive Sera	0	0	0	0	0	0	0
Equines Tested	1	50	102	57	54	49	67
WNV-Positive Equines	0	2	0	0	1	6	8
Mosquito Trap Nights ¹	346	444	2370	1877	1637	2066	1424
Mosquito Pools Tested	47	43	582	1015	915	1225	1314
WNV-Positive Pools	0	0	0	0	2	0	0

¹ Mosquito trap nights are the number mosquito traps set multiplied by the number of days or nights those traps were set. (Example: one EVS light trap set out overnight would be considered one trap night; three EVS traps set out overnight would be considered three trap nights). Selected mosquitoes from these trap nights are pooled together by species and tested for West Nile virus.



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Mosquito Control

Management

Mosquitoes can be managed on an area-wide basis by public agencies that are either components of local health departments or are independent districts organized specifically for mosquito control. There are currently 16 mosquito control districts in Washington. Some are small and have responsibility for mosquito abatement in a few hundred square miles, while the activities of others may encompass one entire county or more. Mosquito control is often accomplished by identifying larval habitat and treating the water with a material that kills the larvae. Many materials currently in use are biological in origin and are highly specific for mosquitoes, with little or no effect on other organisms. On occasion, mosquito abatement agencies may also apply chemical pesticides to kill adult mosquitoes. Adulticiding is usually done, however, only when adult populations become so large that they cause extreme annoyance to many people or when the threat of disease transmission to people is high. Control of irrigation water in agricultural areas to avoid excess runoff is an important mosquito control method, but in recent years, elimination of small bodies of water that can serve as wildlife habitat has ceased to be a mosquito control option because of habitat preservation concerns.

Integrated Pest Management

Mosquito control activities are important to public health, and responsibility for carrying out these programs rests with state and local governments. The current interests in ecology and environmental impact of mosquito control measures, and the increasing problems that have resulted from insecticide resistance emphasize the need for "integrated" control programs. Integrated pest management (IPM) is an ecologically based strategy that relies heavily on natural mortality factors and seeks out control tactics that are compatible with or disrupt these factors as little as possible. Integrated pest management includes the use of pesticides, but only after systematic monitoring of mosquito populations indicates a need. Ideally, an IPM program considers all available control actions, including no action, and evaluates the interaction among various control practices, cultural practices, weather, and habitat structure. This approach uses a combination of resource management techniques to control mosquito populations with decisions based on surveillance. Fish and wildlife specialists and natural resources biologists should be involved in planning control measures whenever delicate ecosystems could be impacted by mosquito control practices.

A good IPM program, featuring surveillance of mosquito populations and disease monitoring, resident education and action to maximize natural controls and minimize mosquito breeding sites, and larviciding when necessary, can control

mosquitoes more effectively while reducing pesticide exposure to humans and the environment. Larvicides should be used where mosquito larvae are shown to be present and not indiscriminately, which is why larviciding is much sounder than adulticiding.

The underlying philosophy of mosquito control is based on the fact that the greatest control impact on mosquito populations will occur when they are concentrated, immobile and accessible. This emphasis focuses on habitat management and controlling the immature stages before the mosquitoes emerge as adults. This policy reduces the need for widespread pesticide application in urban areas.

Mosquito Control Programs

In response to these potential disease-carrying pests, communities organized the first mosquito control programs in the eastern U.S. in the early 1900s.

Eventually, other communities created similar programs throughout the country in areas where mosquito problems occurred and where citizens demanded action by local officials. Modern mosquito control programs in the U.S. are multifaceted and include surveillance, habitat reduction, and a variety of larval and adult mosquito control strategies.

Surveillance methods include studying habitats by air, aerial photographs, and topographic maps, and evaluating larval populations. Mosquito control officials also monitor mosquito traps, biting counts, and complaints and reports from the public. Mosquito control activities are initiated when established mosquito threshold populations are exceeded. Seasonal records are kept in concurrence with weather data to predict mosquito larval occurrence and adult flights. Some mosquito control programs conduct surveillance for diseases harbored by birds, including crows, other wild birds, and sentinel chicken flocks, and for diseases in mosquitoes.

Habitat reduction involves eliminating the habitat or modifying the aquatic habitat to prevent mosquitoes from breeding. This measure includes sanitation measures where artificial containers, including discarded automobile tires, which can become larval mosquito habitats, are collected and properly disposed. Habitat modification may also involve management of impounded water or open marshes to reduce production and survival of the floodwater mosquitoes. If habitat modification is not feasible, biological control using fish may be possible. Contact the Department of Fish and Wildlife before introducing fish into water bodies that are not contained and/or have the potential to reach waters of the state.

Mosquito control officials often apply biological or chemical larvicides, with selective action and moderate residual activity, to aquatic habitats. To have the

maximum impact on the mosquito population, larvicides are applied during those periods when immature stages are concentrated in the breeding sites and before the adult forms emerge and disperse.



Aquatic Mosquito Control

The following are active ingredients which may be used for aquatic mosquito control in Washington. For a guide to the registered products, go to the Washington State Department of Agriculture's Web page at <http://www.kellysolutions.com/WA/pesticideindex.htm>.

Bacillus sphaericus

Bacillus sphaericus (H-5a5b) is a naturally occurring, spore-forming bacterium that is found throughout the world in soil and aquatic environments. It produces a protein endotoxin at the time of sporulation. The toxin is only active against the larval stage and must be ingested and digested before it becomes activated. *B. sphaericus* has the unique property of being able to control mosquito larvae in highly organic aquatic environments such as waste lagoons and storm water catch basins.

First registered for the control of *Culex* mosquitoes, *B. sphaericus* has been expanded to include control of several *Aedes*, *Anopheles*, *Ochlerotatus*, *Psorophora* and *Coquillettidia* species. *Bacillus sphaericus* is not acutely toxic to freshwater and saltwater invertebrates, honey bees, mayfly larvae, does not appear to be harmful to fish and other marine life, and is not toxic to birds on a sub-chronic basis. In tests, *B. sphaericus* was not pathogenic, infective nor toxic in laboratory animals by the oral, dermal, pulmonary or intra-venous routes of exposure. In humans, mild skin and eye irritation can occur with direct contact.

Bacillus sphaericus is available in various formulations such as corn cob granules, water dispersible granules, and water soluble pouches. *Bacillus sphaericus* can offer up to six weeks of control in many habitats because the protoxins and spores can remain suspended in the water column for extended periods and because the bacteria multiply in the dead larvae. Duration of control will depend upon habitat factors such as water depth, flushing, water chemistry and frequency of oviposition to maintain the recycling process.

Bacillus thuringiensis israelensis (Bti)

Bacillus thuringiensis subsp. *israelensis* (*Bti*) is a naturally occurring soil bacterium that can effectively kill mosquitoes during the larvae stage of development in water. *Bti* is an endospore-forming bacterium that is ingested by the actively feeding larvae. When the bacteria *Bti* encysts, it produces a protein crystal toxic to mosquito larvae. Once the bacteria have been ingested, the toxin disrupts the lining of the larvae's intestine causing it to stop eating and die. *Bti* is the primary material used for mosquito control because of its low toxicity to non-target species. *Bti* is highly pathogenic against mosquitoes and black flies, and

has some virulence against certain other Diptera, especially Chironomid midges. *Bti* is highly selective for the first through third instars of mosquito larvae.

Bti has been extensively studied for effects on non-target organisms and environmental consequences of use with no reported adverse effects. It is not toxic to bees. According to several studies, when applied at field application rates, *Bti* has no reported effect on fish and amphibians. Several studies have found no effect on warm-blooded mammals. Labels indicate that direct contact with the products may cause mild eye or skin irritation.

Bti products are available in liquid, pellet, granular, and briquet formulations. The type of *Bti* formulation influences the activity of the product. Generally *Bti* does not persist long after application, with toxicity persisting from 24 hours to over one month when the longer lasting formulations are used. Larval toxicity can depend on the species, its feeding activity and other possible factors such as UV light, water quality, pH, temperature, agitation, and sedimentation. A number of *Bti* products are available for residential use in water bodies, such as lined ornamental ponds, and are typically sold at home and garden-type stores.

Methoprene

Methoprene is a compound that mimics the action of an insect growth-regulating hormone and prevents the normal maturation of insect larvae. Unable to metamorphose, the mosquitoes die in the pupal stage. Methoprene is classified as a biochemical pesticide because it controls mosquito larvae by interfering with the insect's life cycle rather than through direct toxicity. It is effective for controlling first through fourth instars of mosquito larvae. Methoprene comes in various formulations such as briquets, liquid, pellets, water soluble pouches, wettable powder, and granules. Depending on the formulation used and environmental factors, residual control could range from five to 150 days. Some home and garden-type stores have methoprene products that can be used in containerized standing water environments around the home.

Studies indicate that methoprene is of low toxicity and poses little risk to people when used according to label instructions. Methoprene was not shown to have any significant toxicological effects in the standard battery of toxicity studies used to assess human health effects. The pesticide has very low acute oral and inhalation toxicity potential and is not an eye or skin irritant. Methoprene is also of low acute dermal (skin) toxicity and is not a human skin sensitizer.

In laboratory tests, the toxicity of methoprene to birds and fish is low, and it is nontoxic to bees. Field studies involving methoprene have shown that it has no lasting adverse effects on populations of invertebrates or other non-target aquatic organisms when used according to label instructions for mosquito control. Methoprene mosquito control products present minimal acute and chronic risk to freshwater fish, freshwater invertebrates, and estuarine species.

Methoprene is not persistent in the environment. It degrades rapidly in water, being susceptible to transformation by sunlight and microorganisms.

Monomolecular Surface Films

Monomolecular surface film (MSF) is a non-petroleum surface oil that acts as a physicochemical agent by altering the mosquito's habitat. It belongs to the alcohol ethoxylate group of surfactants, which are used in detergent products. Monomolecular surface film disrupts the cohesive properties, which allow mosquitoes to use the water's surface as an interface for breathing or egg-laying. By making the surface "wetter," MSF in effect drowns mosquitoes.

Monomolecular surface film kills larvae and pupae by making it impossible for them to keep their breathing tubes above the water's surface. Mosquitoes that require little or no surface contact for breathing, such as *Coquillettidia* species, require properly timed applications at surface contacting stages – the pupae to emerging adult – for maximum impact. Since MSF kills mosquitoes with a physical mechanism (rather than a toxic mechanism), it is not effective in habitats with persistent unidirectional winds of greater than ten miles per hour, or in areas with very choppy water.

Monomolecular surface film is generally considered non-toxic to most non-target wildlife, however, some species, such as midges that require attachment to the water surface, have been shown to be affected. According to the U.S. Environmental Protection Agency, MSF poses minimal risks to the environment when used as directed. The green tree frog progressed normally from tadpole to adult through several generations after being exposed to a constant film presence for six months. It is not a skin irritant, is only a mild eye irritant on prolonged or repeated contact, and is considered to be non-toxic by animal tests. As with all pesticides, direct contact should be avoided.

The film persistence is dependent on temperature, water flow, amount of bacteria in the water, and the duration and strength of the wind following application. Monomolecular surface film typically persists on the water's surface for 5 – 22 days.

Larvicide oils

Oils, like films, are pesticides used to form a coating on top of water to drown larvae, pupae, and emerging adult mosquitoes. Oils are petroleum or mineral based and are typically used as a product of last resort for the control of mosquito pupae, since this stage does not feed, but does require oxygen. Oils typically persist for 12 to 15 hours and then evaporate within a few days. Larvicide oils, if misapplied, can be toxic to fish and other aquatic organisms. Studies have shown that aquatic invertebrates, amphibians, waterfowl, furbearers and fish may be

deleteriously affected. Consult with the Department of Fish and Wildlife before using larvicide oils.

Chemical larvicides (Restricted or Emergency Use Only)

The application of malathion and temephos, which are organophosphate pesticides, to water are restricted-use larvicides under the Department of Ecology's aquatic mosquito control permit. Temephos is not allowed in lakes, streams, or in the littoral zone of water bodies. Temephos is only allowed in highly polluted and high organic waters with no surface water runoff and may be used in response to the development of pesticide resistance or a public health emergency. Malathion may only be used for control of mosquito larvae under agreement between Washington State Department of Ecology and Washington State Department of Health in response to a public health emergency.



Adult Mosquito Control

The main objective of mosquito control is to decrease the risk of a human outbreak of mosquito-borne disease. This should be primarily accomplished by using integrated pest management principles and:

- Continuing to emphasize reduction in mosquito habitats.
- Larviciding where feasible and practical.
- Using personal protection measures against mosquitoes, especially for the elderly, immune compromised, and those with significant outdoor exposure.

Adulticiding is supplementary to these measures and is a local decision that should be based on the considerations listed (in no particular order) below.

Triggers for Adulticiding

Adulticiding should be considered when there is evidence of mosquito-borne epizootic activity at a level suggesting high risk of human infection (for example, high mosquito infection rates, high dead bird densities, multiple positive mosquito species including key bridge vectors, horse or mammal cases indicating escalating epizootic transmission, or a human case with evidence of epizootic activity) and abundant adult vectors. In general, the finding of a positive bird or mosquito pool does not by itself constitute evidence of an imminent threat to human health and warrant mosquito adulticiding, but, intensifying surveillance would be recommended to help identify any increasing threats.

Where to Adulticide

Another consideration is the terrain in the proposed spraying area. If there is substantial vegetation bordering the roads, ground spraying with trucks may not provide adequate coverage. Dense vegetation associated with roadside trees, shrubs, or hedges can interfere with truck-mounted insecticide applications. In situations such as these, counties that choose to spray may wish to consider application of pesticides using backpack sprayers, or altering their route so that trucks can more efficiently apply the spray. Aerial application may be considered when all other methods of application are inadequate and/or inefficient. Aerial spraying should be limited to the immediate area where the vector population has been documented to exist through vector surveillance and to adjacent areas considered at risk for imminent disease transmission.

You can contact the Washington State Department of Ecology for guidance in situations involving spraying near water or with applications that might be hindered by vegetation.

Human Population Density

The population density in an area where there is evidence of intense epizootic activity should also be taken into consideration. If the area is rural and there are few people, the cost and potential risks of human transmission may not justify the use of adulticides. If the area is heavily populated, you have stronger indications for considering adult mosquito control, since the goal of spraying is to minimize the risk of a human outbreak of mosquito-borne disease.

Mosquito Population

Information from mosquito surveillance can be helpful in determining when to conduct mosquito control, and in monitoring the effectiveness of control activities. While all mosquitoes do not need to be tested specifically for the presence of viruses, those that are tested can provide valuable information regarding control decisions. Surveillance efforts to detect virus in birds are typically easier to conduct than similar efforts to detect virus in mosquitoes. What may be more important than testing mosquitoes is knowing the abundance and species of the vector population in the locality. The best way to do this is by mosquito trapping. Systematic mosquito trapping, however, requires trained staff and is time intensive. For localities without this capacity, there are other potential sources of information on mosquito activity. Staff can visually inspect the area where a positive bird was found, or around human population centers for habitats likely to be conducive to mosquito breeding. Staff can also personally observe mosquito activity.

Lag Time

It is important to look at the dates that the positive surveillance specimens (mosquitoes, birds, and/or mammals) were collected. In most cases the positive specimens will have been collected about one week before. In the time between the date the specimen was collected and the date when the test results are complete, circumstances may have occurred which would alter a decision to use adulticides. For example, a county may have sprayed since the collection date, a weather event may have adversely affected mosquitoes, or mosquito habitat may have been modified resulting in a reduced need to spray.

West Nile Surveillance Results Over Time

WNV surveillance information may be monitored by county or even smaller jurisdictions, such as towns, over time, to determine what is happening with the outbreak. For example, if there has been a consistently good system for recording dead crow sightings, and the number of dead crow sightings drops for several weeks in a row after adulticiding, that may indicate that the previous spraying has killed off a large enough number of mosquitoes that the transmission to crows is

not continuing. Such analyses should not be graphed by day (because of day-to-day instability in reporting), but analysis by week should be helpful.

Local Perspectives on Adulticiding

Different communities have varying perspectives on the benefits of mosquito control. These should be taken into account in the decision whether or not to use adulticides. This can be difficult, as people can have strong opinions on both sides of the issue. All pesticides used for mosquito control are United States Environmental Protection Agency approved and persons using restricted use pesticides must also be tested and licensed by the Washington State Department of Agriculture. For further information about toxicity of the common pesticides used for mosquito control, go to the United States Environmental Protection Agency Web site at <http://www.epa.gov/pesticides/health/mosquitoes/>.

Before events force a decision regarding whether to use adulticides or not, local health jurisdictions should assess their ability to conduct or assist in the coordination of adult mosquito control if needed. Some of the questions to consider include:

- Who in the community is qualified to apply mosquito control pesticides?
- What equipment is available in the community to conduct adult mosquito control?
- Where are the amplification and vector species of mosquitoes located and at what times of the year do they appear?
- Can local mosquito surveillance be improved to reduce the cost of mosquito control?
- Will the community need to contract with commercial businesses or others for public health mosquito control?
- Who will be the lead for public health mosquito control?
- How will the community be involved and informed during the decision process?
- How can we evaluate the effectiveness of adult mosquito control?

The decision regarding adulticiding is basically a risk assessment: whether or not you, as a community, believe the risk of contracting a mosquito-borne disease is greater than the risk from applying pesticides for mosquito control. It is also a cost assessment where you must take into account medical costs, life years lost (for fatalities), costs of spraying campaigns, etc. As with any decision about access to health prevention and care, many factors must be considered.



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Appendix F – Public Information

Early Season Messages [105](#)

High Season Messages [107](#)

End of Season Messages [109](#)

News Release Early Season West Nile Virus Message [111](#)

News Release West Nile Virus Detected [113](#)

News Release Human West Nile Virus [115](#)



Early Season Messages

March – June

- Mosquitoes are starting to appear.
- Mosquitoes may carry disease.
- Your local health agency (or mosquito control district, if applicable) are prepared to conduct mosquito-borne disease monitoring and coordinate control efforts.
- Find and eliminate mosquito development sites. Remove standing water around your home and your place of business.
- Mosquitoes can develop in unlikely locations – a jar lid filled with water, clogged rain gutters. Discarded tires are a particular problem.
- Personal protection measures include avoiding mosquito bites by using effective repellent, wearing protective clothing, being aware of prime mosquito biting hours, and repairing/installing household screens.
- Dead crows, magpies, ravens, jays, and raptors could be a sign of WNV. Be sure to report them to your local health agency. Not all dead birds will be picked up for testing.
- Most people who have been bitten by a WNV-infected mosquito won't show any signs of illness. Some people may develop flu-like symptoms that go away without treatment. However, some people may have lingering symptoms of headache or fatigue. In a small number of cases the virus can cause serious illness including fever and inflammation of the brain or spinal cord. People over age 50 and those with weakened immune systems have the highest risk for serious illness.



High Season Messages

July – September

- Continue early season messages with emphasis on bite prevention.
- Mosquito populations are at their peak.
- Continue to find and eliminate mosquito development sites. Eliminating these water body sites around the home will help reduce mosquito populations, lowering your risk of being bitten, and may help reduce the need for additional control measures later on.
- We expect to detect WNV in our state this season. By testing dead birds, mosquitoes, and symptomatic horses and people, we can determine where the virus is present. Monitoring for the virus provides our local community an early warning so we can target education and control activities.
- If spraying insecticide that targets adult mosquitoes (adulticiding) becomes necessary, here are the facts you need (include information about products being used, their safety, and environmental concerns).



End of Season Messages

October – November

- Mosquito populations are declining.
- Monitoring and testing for mosquito-borne diseases is ending.
- Summarize the season's virus activity.
- Plans for next year's mosquito season.



News Release Early Season West Nile Virus Message

Sample from the Washington State Department of Health

For immediate release: Date

Contacts: Name, title, phone number
 Name, title, phone number

Environmental monitoring for West Nile virus resumes around the state

West Nile virus infection was found in birds, horses and humans in the state last year

OLYMPIA - West Nile virus is establishing itself in Washington. In an effort to provide local communities an early warning of the disease, dead birds and mosquitoes are being tested for the virus.

In (previous year), West Nile virus was detected in ___ people, ___ dead birds, ___ horses, and ___ mosquito pools in the state. If Washington follows the trend seen in other states, there could be more human infections in the state this season.

“Mosquitoes samples and dead birds are often the first warning that the disease has arrived in the area,” said (name, title, office). “Local communities can then increase their monitoring and mosquito control, and let residents know how to protect themselves.”

Crows, jays, magpies, ravens, and raptors (such as hawks and owls) are especially susceptible to dying from West Nile virus – making them ideal candidates for testing. Whenever these types of birds are found dead, it should be reported to the local health agency in the area.

Mosquitoes that can pick-up West Nile virus from birds and spread the disease to people are also being targeted for testing. Several local health agencies, mosquito control districts, and volunteer groups are trapping adult mosquitoes for testing.

The best protection against West Nile virus is to avoid mosquito bites. Stay indoors around dusk and dawn when mosquitoes are most active, cover exposed skin, install window screens, and use an effective mosquito repellent. Reduce mosquito breeding habitat by turning over old buckets or cans, emptying water from old tires, and change water in birdbaths, wading pools, fountains, and water troughs once or twice a week.

Most people bitten by a mosquito carrying West Nile virus won't become ill. Some may have mild to severe flu-like symptoms and a small number of people may develop a serious neurological disease. Anyone can contract West Nile virus

and become ill, but people over 50 years old and those with compromised immune systems are at greater risk of serious illness.

The Department of Health West Nile virus information line 1.866.78VIRUS (1.866.788.4787) and the agency's West Nile virus Web site (www.doh.wa.gov/WNV) are excellent resources for anyone who wants to learn more.



News Release West Nile Virus Detected

Sample from the Washington State Department of Health

For immediate release: Date

Contacts: Name, title, phone number
Name, title, phone number

Dead bird from _____ County tests positive for West Nile virus

OLYMPIA - A dead bird from _____ County has tested positive for West Nile virus. According to the Washington State Department of Health, the crow was collected in early August and sent to the Washington Animal Disease Diagnostic Laboratory at Washington State University for testing. It's the first West Nile virus-positive dead bird in our state this year. Last year the virus was detected in ___ people, ___ horses, ___ dead birds, and ___ mosquito pools from ___ counties around the state.

“Testing for West Nile virus in dead birds, mosquitoes and ill horses helps show where the disease is active,” said (name, title, office). “These tests show West Nile is active in parts of Washington and serves as a reminder for people to avoid mosquito bites.”

The crow was collected and submitted by the _____ Health District as part of the state's monitoring program. Many local and state agencies as well as private volunteer groups are participating in the monitoring. The program has submitted nearly ___ dead birds for testing this season and this is the only positive result, so far. People are encouraged to continue reporting dead birds, especially crows, jays, magpies, and ravens, to local health agencies for possible collection and testing.

“As West Nile virus continues to establish itself in our state, it's possible we will see more cases in animals and in people this year,” said (name). “Local governments should be implementing education, surveillance, and control strategies to help combat the spread of West Nile virus. Local residents can take precautions to avoid mosquito bites and limit mosquito breeding around their homes.”

Simple measures can reduce the risk of exposure to mosquitoes that transmit West Nile virus. Avoiding mosquito bites is the key:

Schedule outdoor activities away from dusk and dawn when mosquitoes are most active.

Wear long-sleeved shirts and long pants when in mosquito infested areas.

Use an effective mosquito repellent and follow label instructions. Repellents containing DEET, picaridin, oil of lemon eucalyptus, PMD – the synthesized version of oil of lemon eucalyptus, or IR3535 have been shown to be effective.

Make sure the screens on all doors and windows are working properly.

Eliminate mosquito breeding by emptying out anything that holds standing water such as old tires, buckets, plastic tarps, and clogged gutters. Change the water in birdbaths, fountains, wading pools, and animal troughs twice a week.

West Nile virus is primarily a bird disease. Mosquitoes become infected by feeding on an infected bird and can pass the virus to humans, horses or other hosts.

“Most people infected with West Nile virus don’t have any symptoms but in a few cases, the virus can cause serious illness including inflammation of the brain or spinal cord. Anyone can get West Nile but people over age 50 have the highest risk for serious illness,” said (name, title, office). “There’s no specific treatment for West Nile virus infection.”

More information on West Nile virus (www.doh.wa.gov/WNV) is available on the Department of Health Web site and online from The Centers for Disease Control and Prevention (www.cdc.gov/ncidod/dvbid/westnile/). (Name) of the _____ Health District is available at (phone number) to answer questions about local West Nile virus monitoring and other local environmental health issues.



News Release Human West Nile Virus

Sample from the Washington State Department of Health

For immediate release: Date

Contacts: Name, title, phone number
Name, title, phone number

_____ County man is state's first confirmed case of West Nile virus this year

OLYMPIA – A _ County man in his forties was confirmed as the state's first human infection with West Nile virus this year. Blood tests at the state Public Health Laboratories were positive, so samples were sent to the Centers for Disease Control and Prevention (CDC), which confirmed the positive result today. The man developed mild symptoms — a fever and rash — in August, sought medical attention, and has recovered. The disease is not spread person-to-person so he is not a threat to public health.

“Most people who are infected with West Nile virus don't ever get sick, yet it can be a very serious disease for a small number of people, especially those older than 50 years of age,” said (name, title and office). “Avoiding mosquito bites is the best prevention for not getting a West Nile virus infection.”

People can avoid mosquito bites by staying indoors around dawn and dusk when mosquitoes are most active; making sure that door and window screens are in good working condition; and using an effective mosquito repellent when outdoors in areas where mosquitoes are active. It's also important to reduce mosquito habitat around the home. Emptying or getting rid of water from buckets, cans, and old tires, and frequently changing water in birdbaths, fountains, wading pools, and water troughs helps eliminate the small puddles of water in which many mosquito species breed.

West Nile infection can cause a wide spectrum of illness from no symptoms at all to a rash and headache to serious neurological disease including encephalitis (inflammation of the brain) or meningitis (inflammation of the lining of the spinal cord and brain). People over age 50 have the highest risk for serious illness.

West Nile virus is primarily a bird disease. Mosquitoes become infected by feeding on infected birds, and then pass the virus to uninfected birds, humans, horses or other hosts. Crows, ravens, magpies, jays, and raptors are especially susceptible to dying from the virus. The Department of Health is monitoring for the presence of West Nile virus by testing mosquito samples, dead birds, and ill horses. So far in (year), positive West Nile virus cases have been identified in

___ horses (___ tested), ___ dead birds (___ tested), and ___ mosquito pools (___ tested).

The Department of Health West Nile virus information line 1.866.78VIRUS (1.866.788.4787) and the agency's West Nile virus Web site (www.doh.wa.gov/WNV) are excellent resources for anyone who wants to learn more.

The Department of Health is working with local health partners as well as other state agencies on West Nile virus monitoring, prevention, and control.



Appendix G - Regulations

Referenced RCWs [117](#)

Glossary [119](#)



Referenced RCWs

The following web links will take you to the RCWs that have been referenced in this guidance:

- RCW 70.05.060
<http://apps.leg.wa.gov/RCW/default.aspx?cite=70.05.060>
- RCW 70.05.070
<http://apps.leg.wa.gov/RCW/default.aspx?cite=70.05.070>
- Washington Constitution Article XI, Section II
<http://www.leg.wa.gov/LawsAndAgencyRules/constitution.htm>
- Title 35 RCW
<http://apps.leg.wa.gov/rcw/default.aspx?Cite=35>
- Title 35A RCW
<http://apps.leg.wa.gov/rcw/default.aspx?Cite=35A>
- Title 36 RCW
<http://apps.leg.wa.gov/rcw/default.aspx?Cite=36>
- RCW 70.22.060
<http://apps.leg.wa.gov/RCW/default.aspx?cite=70.22.060>
- RCW Chapter 17.28
<http://apps.leg.wa.gov/RCW/default.aspx?cite=17.28>
- RCW 70.22.020
<http://apps.leg.wa.gov/RCW/default.aspx?cite=70.22.020>
- RCW 70.22.030
<http://apps.leg.wa.gov/RCW/default.aspx?cite=70.22.030>



Glossary

Adulticide: Pesticide used to control insects at the adult stage of their development.

Amplification: A general increase in the number of parasites or pathogens in a given area.

Amplifying host: A parasite host in which the number of parasites or pathogens increases and therefore the number of infected vectors feeding on that host also increases.

Antigen: A protein or carbohydrate capable of stimulating an immune response. An **epitope** or **antigenic determinant** is a molecular region on the surface of an antigen capable of eliciting an immune response and of combining with the specific antibody produced by such a response.

Arbovirus: Any of various ribonucleic acid (RNA) viruses transmitted by arthropods. Arbovirus is not a taxonomic group, but derived from the term **arthropod-borne virus**.

Arthropod: Invertebrate animals in the phylum Arthropoda, a group that have a segmented body, jointed appendages, a usually chitinous exoskeleton molted at intervals, and a dorsal anterior brain connected to a ventral chain of ganglia. Includes insects, arachnids, and crustaceans.

Bridge Vector: This is a species of arthropod that acquires a disease-causing agent from an infected wild animal and subsequently transmits the agent to a human. For example, *Culex salinarius* is a mosquito whose females feed readily on both birds from which they may acquire West Nile virus and on mammals to which they may transmit the virus during the taking of a subsequent blood meal.

Carrier: A bearer and transmitter of a causative agent of an infectious disease; especially one who carries the causative agent of a disease systemically but is immune to it.

Diptera: A large group (Order) of insects that are abundant almost everywhere and include mosquitoes, flies, gnats, and midges. Diptera are distinguished from other Orders of insects by having only one pair of wings, with the second pair instead reduced to small knobbed structures called "halteres" that aid in maintaining equilibrium. Some diptera are vectors of diseases such as malaria, yellow fever, filariasis, and the other arboviruses, while others are important pollinators, pest predators, and parasites. Diptera undergo complete metamorphosis, which means that they change form during development (see larvae and pupae).

Encephalitis: An inflammation of the brain that can be caused by viruses and bacteria, including viruses transmitted by mosquitoes.

Endemic: Belonging or native to a particular people or country and thus continuously present at the expected frequency of occurrence; restricted or peculiar to a locality or region (endemic diseases; an endemic species).

Enzootic: Referring to animal diseases that are peculiar to or constantly present in a locality.

Epidemic: Affecting or tending to affect a disproportionately large number of individuals within a population, community or region at the same time. (i.e., at a higher than expected frequency). Used to refer to diseases that are not consistently present in an area, and which are brought in from the outside or a temporary increase in the number of cases of an endemic disease.

Epidemiology: A branch of medical science that deals with the incidence, distribution, and control of disease in a population; or the sum of the factors controlling the presence or absence of a disease or pathogen.

Gonotrophic cycle: The cycle the female mosquito goes through that begins with the search for a host and the taking of a blood meal, involves the maturation of a batch of oocytes (eggs), and ends with oviposition.

Gravid: Distended with or full of eggs, as in “mosquito trap for gravid females” also called “gravid trap.”

Host: A living animal or plant affording subsistence or lodgment to a parasite. Parasites are organisms that are metabolically dependent upon the host.

Instar: The insect between successive molts, the first instar being between hatching and the first molt.

Larva (plural **larvae**): the immature stages between the egg and the pupa, of an insect with complete metamorphosis. The form of the insect during the larval stage differs radically from the adult.

Larvicide: Pesticide used to control insects at the larval stage of their development.

Morbidity: The relative incidence of disease.

Multivoltine: Having several broods in a season, as in “multivoltine species of mosquito.”

Oviposit: To lay or deposit eggs.

Predation: A mode of life in which food is primarily obtained by the killing and consuming of animals.

Predaceous: Living by preying on other animals: predatory.

Pupa, (plural **pupae**): The immature stage between the larva and adult, of an insect with complete metamorphosis.

Recrudescence: A new outbreak after a period of abatement or inactivity.

Recrudescence: To break out or become active again. (e.g., some scientists hypothesize that West Nile virus may be a chronic infection in birds that recrudesces during times of stress for the bird, such as migration, mating, and when establishing territory).

Reservoir: A population or group of populations of vertebrate or invertebrate hosts in which the pathogen is endemic (i.e., permanently maintained). Although human populations can form reservoirs of this kind, the concept is usually applied to non-human populations from which the pathogen periodically escapes, causing individual infections or epidemics in humans or epizootics in other animals.

Sequela (plural **Sequelae**): An after effect of disease or injury. A secondary result.

Seroconversion: The production of antibodies in response to an antigen.

Seropositive: Having or being a positive serum reaction, especially in a test for the presence of an antibody.

Seroprevalence survey: A standard tool used by public health officials. “Sero-” (from the Latin serum) refers to testing of blood for antibodies to an infectious organism; “prevalence” refers to the percentage of people with a particular characteristic at a given point in time. A "West Nile Virus Seroprevalence Survey" is a survey to determine the percentage of persons with antibodies to West Nile virus, at a given point in time, within the geographic area sampled.

Teneral: Recently molted adult mosquitoes, usually still pale and soft-bodied individuals.

Transovarial: With regard to West Nile virus, used in phrase “transovarial transmission” and refers to capability of an infected mosquito to pass the virus to offspring, a process also known as vertical transmission. Infection in male mosquitoes would be attributed to transovarial transmission since male mosquitoes do not bite to get a blood meal.

Trap night: One trap operated for one night. Three traps operated for one night each would equal three trap-nights.

Vector: Carrier of a pathogen from one host to another.