



Surveillance and Control of Selected Arthropod-borne Diseases in Florida

2003



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Purpose

This publication establishes guidelines for detecting and monitoring St Louis Encephalitis, West Nile and other arthropod-borne diseases and minimizing the risk of human infection. This manual identifies functions and prescribes responsibilities which will assure that appropriate prevention and control methods are initiated promptly and effectively. Please address comments to Dr. Carina Blackmore, Bureau of Community Environmental Health, 4052 Bald Cypress Way, Bin A-08, Tallahassee, Florida 32399-1720, (850) 245-4299, FAX (850) 922-8473.

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Chapter 1

General Information -- Selected Arthropod-borne Diseases

I. Arboviruses

Arthropod-borne viruses, *i.e.* "arboviruses", are viruses that are maintained in nature through transmission between susceptible animal hosts by blood-feeding arthropods (e.g., mosquitoes and ticks). 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 in the vector (e.g., the virus is transmitted from the female to the offspring).

Arboviral encephalitides have a [global distribution](#). Arboviral agents of encephalitis in the United States include: St. Louis encephalitis (SLE) virus, West Nile (WN) virus, eastern equine encephalomyelitis (EEE) virus, western equine encephalomyelitis (WEE) virus, Venezuelan equine encephalomyelitis (VEE) virus, Everglades virus (EVE), California encephalitis (CE) virus and LaCrosse (LAC) encephalitis virus, all of which are transmitted by mosquitoes. Another virus, Powassan, a tick borne virus, is a minor cause of encephalitis in the northern United States. Most cases of arboviral encephalitis occur from June through September, when arthropods are most active. In Florida, where arthropods 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 syndrome. Onset may be insidious or sudden with fever, headache, myalgias, malaise and occasionally prostration. Infection may, however, lead to encephalitis, with a fatal outcome or permanent neurologic sequelae. Fortunately, only a small proportion of infected people progress to having encephalitis.

Laboratory criteria for arboviral encephalitis diagnosis include: a four-fold or greater change in serum antibody titer between acute and convalescent samples; virus isolation or viral antigen identified in tissue, blood or cerebrospinal fluid (CSF); or specific immunoglobulin M (IgM) in blood or CSF identified by enzyme immunoassay (EIA) antibody confirmed by demonstration of IgG via another serologic assay (e.g., hemagglutination-inhibition (HI) or neutralization test).

Because the arboviral encephalitides are viral diseases, antibiotics are not helpful for treatment and the effectiveness of antiviral agents has not been shown. Treatment is supportive, attempting to deal with problems such as swelling of the brain, loss of automatic breathing activity and other treatable complications like bacterial pneumonia. There are no commercially available human vaccines for these U.S. diseases. A vaccine is available for horses and ruminants against EEE, WEE and Venezuelan equine encephalitis (VEE). A horse vaccine against WN virus has been on the market since 2001.

Arboviral encephalitis can be prevented through personal and community protective measures. Personal protective measures include reducing time outdoors particularly in early morning and evening hours, wearing long pants and long-sleeved shirts, applying mosquito repellent to exposed skin areas and maintaining screens/doors. Residual insecticide applications on and around screen doors give added protection. Community preventive measures include reducing mosquito-breeding sites around residences (e.g., dumping

water collected in flowerpots, wading pools and buckets and removing/destroying discarded tires) and may include the use of insecticides (larvicides and adulticides) to kill mosquitoes.

Several local, state and federal agencies are involved with the surveillance and control of arboviral diseases. Mosquito-borne encephalitis surveillance activities include evaluating mosquito populations, sentinel chickens, wild birds, and other animal cases to detect the risk of disease before it occurs in people and to intervene to reduce that risk substantially. Rapid diagnostic techniques used in threat recognition can shorten public health response time and reduce the geographic spread of infected vectors, and thereby, the cost of containing them.

The surveillance required to detect risk is being increasingly refined by the potential utilization of new technologies which allow for rapid identification of dangerous viruses in bird mosquito populations. Virus isolation and identification are useful in defining viral agents in mosquito vectors. While virus isolation still depends upon growth of virus in cell culture or neonatal mice, virus identification has been greatly facilitated by the availability of virus-specific genomic sequence information for use in polymerase chain reaction (PCR) assays and monoclonal antibodies (MAbs) for use in IFA and ELISA assays. MAbs with avidities sufficiently high to allow for specific binding to virus antigens in a complex protein mixture (e.g., mosquito pool suspensions) have also enhanced the ability to rapidly identify virus agents *in situ*.

A. St. Louis Encephalitis (SLE)

St Louis encephalitis virus, a flavivirus, was prior to the introduction of WN virus the most common mosquito-transmitted human pathogen in the U.S. During the summer season, SLE virus is maintained in a mosquito-bird cycle, with periodic amplification by birds and *Culex* mosquitoes. In Florida, the principal vector is *Cx. nigripalpus*, a ubiquitous species found throughout central and south Florida.

Infection with SLE virus results in inapparent infection in a variety of birds and mammals with a resultant period of viremia that lasts a matter of days. Humans represent an incidental, dead-end host. The estimated incubation range is four to 21 days. The clinical spectrum of human SLE infection includes inapparent infection, mild illness (fever with headache), aseptic meningitis, and encephalitis that can progress to coma and death. Less than 1% of SLE viral infections in people are clinically apparent and the vast majority of infections remain undiagnosed. Encephalitis, especially that progressing to coma and death, is more common in the elderly. The case fatality rate in Florida SLE epidemics has ranged from 4 to 30 percent. Deaths were almost exclusively among people age 50 and older.

The first recognized SLE outbreak occurred in St. Louis, Missouri in 1933. Since then, many SLE epidemics have been documented in North America with the vector species varying by region. In Florida, SLE outbreaks were documented in 1959 (N=68), 1961 (N=25), 1962 (N=222), 1977 (N=110), 1980 (N=10), 1990 (N=223), 1993 (N=8) and 1997(N=9). The epicenter of the outbreaks was the Tampa Bay area for all years but 1977 and 1990. In 1980, six sporadic cases of SLE were reported from counties around Tampa Bay (Pinellas, Hillsborough, Pasco, Manatee and Sarasota). In addition, four cases were reported from residents of Fort Walton Beach in Okaloosa County; this incident was particularly interesting in that human cases of SLE had never before been documented in the panhandle of Florida. These cases also occurred between July 10 and August 2, much earlier than expected.

These outbreaks stimulated the establishment of research into mosquito-borne diseases and mosquito control activities including two arbovirus research facilities (in Tampa and Vero Beach). The most widely used surveillance technique in Florida has been the use of chicken sentinel flocks, and these have been maintained in several Florida counties.

B. Eastern Equine Encephalomyelitis (EEE)

Eastern equine encephalomyelitis virus is an [*alphavirus*](#) that was first identified in the 1930's and currently occurs in focal locations of the United States. EEE virus occurs in natural cycles involving birds and

Culiseta melanura in freshwater swampy areas with a peak of activity between May and August. Where the virus resides or how it survives in the winter is unknown. Migratory birds may introduce it in the spring, or it may remain dormant in some yet undiscovered part of its life cycle. With the onset of spring, the virus reappears in the birds (native bird species do not seem to be affected by the virus) and mosquitoes of the swamp. In this usual cycle of transmission, virus does not escape from these areas because the mosquito involved prefers to feed upon birds and does not usually bite humans or other mammals.

For reasons not fully understood, the virus may escape from enzootic foci in swamp areas in birds or bridge vectors such as *Coquilleltidia perturbans*, *Ochlerotatus atlanticus*, *Cx. nigripalpus*, *Cx. perturbans*, *Cx. quinquefasciatus*, *Oc. sollicitans*, and *Aedes vexans*. These species feed on both birds and mammals and can transmit the virus and cause disease in people, horses, puppies and some birds such as pheasants, quail, ostriches and emus. While small outbreaks of human disease have occurred in the United States, equine epizootics can be a common occurrence in unvaccinated populations because horses are outdoors and attract hordes of biting mosquitoes. Human cases may be preceded by those in horses; therefore, horse cases may be used as a potential surveillance tool.

It takes from 4-10 days after the bite of an infected mosquito for an individual to develop symptoms of EEE. These symptoms begin with a sudden onset of fever, general muscle pains, and a headache of increasing severity. Many individuals will progress to more severe symptoms such as seizures and coma. Approximately one-third of all people with clinical encephalitis caused by EEE will die from the disease and, of those who recover, many will suffer permanent brain damage requiring long-term medical care.

Human and equine cases occur within five miles of *Cs. melanura*-producing swamps. All evidence indicates that human EEE does **not** have epidemic potential in Florida. Continuous surveillance for the past forty years (1957-97) has documented only 62 sporadic cases in people (average 1.6 cases per year; range 0-5). Additionally, avian serosurveillance does not appear to be as useful as for predicting SLE cases in people. Still, health officials can maintain surveillance for EEE virus activity with the aid of mosquito control officials. If the level of activity is sufficiently high, mosquito control and personal protection are recommended to reduce the risk to humans.

Whereas *Cs. melanura* is distributed statewide, human (and equine) cases have predominantly been in areas north of Lake Okeechobee. In particular, there have been clusters of cases in seven areas: Escambia County; Walton-Holmes-Jackson counties; Duval County; Alachua-Marion counties; Leon-Jefferson-Madison counties; the lower St. Johns area of Volusia, Flagler, Putnam and Clay counties; and the Green Swamp region of Lake, Orange, Pasco, Polk, Osceola, Pinellas, Hillsborough and Manatee counties.

C. West Nile (WN) virus

The WN virus outbreak in the northeastern U.S. in the summer and fall of 1999 represented the first known incursion of this exotic arbovirus into the U.S. Since then the virus has spread and by the end of 2002, it had been detected in 44 states and more than 4,200 human cases had been confirmed. WN virus was first detected in Florida in July, 2001. Twelve human cases were reported in the state that year. In 2002, 28 human cases of WN meningoencephalitis and seven cases of WN fever were detected in Florida. Included among these cases were two individuals who acquired their infections via organ transplants and one person who became infected from a blood transfusion. WN virus activity has been detected in all 67 Florida counties. In 2001, the epicenter of the WN virus outbreak was in the north-central part of the state. During WN virus second year in Florida the most intense virus activity was detected in northwestern and central Florida counties.

Like SLE virus, which is closely related to WN virus, the natural cycle of WN virus appears to involve *Culex* mosquitoes and wild birds. However, unlike SLE virus, WN virus causes high rates of mortality in certain families of birds, especially in corvids. It is also pathogenic for horses. More than 900 cases of equine WN meningo-encephalitis were confirmed in Florida in 2001 and 2002.

Because SLE and WN viruses are antigenically related, cross-reactions are observed with some serologic tests. Plaque reduction neutralization testing (PRNT) done to distinguish the two viruses is done at the Department of Health Laboratory, Tampa.

D. Other Arboviral Encephalitides

Other arboviral encephalitides of minor public health significance that occur in Florida are WEE, EVE and Keystone (*Bunyaviridae*; California group). To date no reported human cases of WEE have been acquired in Florida. While serologic evidence of EVE virus infection has been documented in south Florida, only three clinical cases have ever been identified, two near Homestead and Florida City in Dade County (1968 and 1971) and one near Vero Beach (1968). The only recorded human case of Keystone virus occurred in a young child from Sarasota in 1964.

II. Lyme Disease

Lyme disease (LD) is caused by a spirochete bacterium known as *Borrelia burgdorferi*. The disease derives its name from Lyme, Connecticut, where cases of unusual juvenile arthritis were first studied in the early 1970s, and the agent later identified as being transmitted through infected ticks. The black-legged tick, *Ixodes scapularis*, is the suspected vector in the southeast, although *Amblyomma* ticks may also be important. Ticks acquire the spirochete by feeding on wild mice and other rodents that serve as the primary reservoir of infection. The spirochete thrives and multiplies within certain species of ticks and during subsequent feeding is transmitted to other hosts. The presence of larger animals, such as deer, is known to be important in maintaining large tick populations in an endemic area.

If bitten by an infected tick (often nymphal stages), most people will experience a red, "bull's eye" rash (erythema migrans or EM) three to 30 days later. The rash does not always occur at the site of the bite, but may appear at the armpit, groin or back of the knee. Other symptoms of LD include fatigue, neck stiffness, muscle aches and flu-like symptoms such as headaches, chills, fever or dizziness. Later stage symptoms may not appear until weeks, months, or years after the tick bite and can include neurologic, musculoskeletal and cardiac problems. Unless treated with antibiotics within the first few months of infection, LD can become a highly debilitating, but rarely fatal illness capable of producing symptoms in both humans and domestic animals (i.e., dogs, cats, horses and cattle).

Serologic tests available for LD diagnostics include IFA, EIA, and immunoblotting. Poorly standardized tests must be interpreted cautiously. False-positive reactions may result from cross-reacting IFA and EIA antibodies in patients with syphilis, leptospirosis, Rocky Mountain spotted fever, infectious mononucleosis, lupus or rheumatoid arthritis. Antibiotic treatment during the early stages of the disease may limit the antibody response, however serum samples from persons with disseminated or late-stage LD almost always have a strong IgG reactivity with a typical banding pattern to *Borrelia burgdorferi* antigens by Western immunoblotting. Skin biopsies of the EM lesion may yield *Borrelia* organisms.

LD occurs throughout the continental US with highest incidence in foci in the northeastern, northcentral, mid-Atlantic and northern Pacific regions. LD case reporting has risen substantially over the last decade, at least in part, because of greater awareness of the illness. Some are concerned about overdiagnosis of LD and the resulting, inappropriate treatment.

LD occurs only sporadically in the southern states. In Florida, most people with LD acquired their infection in the northeast. In the five-year period 1998-2002, an annual average of 24 cases without a travel history outside of the state were reported to the State Health Office. Physicians are reminded that diagnosis of LD is required to be reported.

III. Rocky Mountain Spotted Fever

Rocky Mountain Spotted Fever (RMSF) was first recorded in 1896 when human cases were described in Idaho. Unlike its name implies, RMSF, is now rarely reported from the Rocky Mountain regions. Synonyms for the disease include tick-borne typhus or tick typhus.

Disease is caused by infection with the intracellular coccobacillary bacteria, *Rickettsia rickettsii*, following tick exposure. Ninety percent of the thousand rickettsial disease cases that occur annually in the United States are RMSF. The principal tick vectors in Florida are probably the dog tick (*Dermacentor variabilis*) and the Lone Star tick (*Amblyomma americanum*). A tick bite may or may not be apparent and malaise, muscle pain, headache and chills are not uncommon. In most cases a mild febrile illness develops after an incubation period of a few days to 2 weeks. About one-half of the cases also develop a maculopapular rash that appears first on the extremities and spreads to the trunk.

In the early 1970's an increase in cases of reported RMSF in Florida paralleled national trends. Prior to that time, this disease was diagnosed infrequently in the state with only 25 confirmed cases reported in the 31 year period between 1942 and 1972. Documentation of the travel history on these cases indicated that only two may have been acquired in Florida. Between 1973 and 1976 the HRS Division of Health investigated 15 confirmed cases and found that 12 (80%) had no travel history outside of the state. In 1985, Sacks and Janowski reviewed the histories of 49 confirmed RMSF cases reported in Florida between 1973 and 1983. Analysis of the 25 cases believed to have been acquired in the state showed that RMSF infections tended to occur during the warmer months, March through November, with a peak in August. Cases ranged in age from 2 to 72 years; the median age was 24 years. Males accounted for 68% and whites 88% and exposure was linked to 21 different counties. Sixteen of the cases (64%) had a known tick bite or attachment, three (12%) had been deticking animals and six (24%) had no known tick exposure. However, those in the latter group had a history of contact with dogs or outdoor activities.

Between 1998 and 2002, 31 cases of RMSF were reported in Florida. Historically, more cases have occurred in northern counties. Still, it appears that there is a potential risk for RMSF wherever people are exposed to ticks.

IV. Human Ehrlichiosis

Bacteria in the genus *Ehrlichia* cause two recently recognized and potentially fatal diseases. *E. chaffeensis*, discovered in 1987, causes human monocytic ehrlichiosis (HME). A species of *Ehrlichia* closely related to *E. phagocytophilia* and *E. equi* causes human granulocytic ehrlichiosis (HGE). Nonspecific clinical findings make these diseases difficult to diagnose. They may account for many cases of unexplained tick-associated fevers of unknown origin -- for example, some illnesses misdiagnosed as LD.

HME has been identified in over 1,000 patients in the United States, Europe, and Africa. Most cases in the U.S. occur in adults from rural areas of southern states between April and September. The most likely tick vector is *Amblyomma americanum*. The spectrum of illness ranges from asymptomatic to fatal. Most cases have a nonspecific febrile illness without rash, with over 60% hospitalized. About 15% have severe infections, including renal failure, disseminated intravascular coagulopathy, seizures, and coma, and 2 to 3% die. Laboratory findings often include leukopenia, thrombocytopenia, and elevated serum hepatic enzymes. Early diagnosis is rare because morulae of *E. chaffeensis* are seldom found in peripheral blood, seroconversion does not occur until convalescence and in vitro cultivation has been accomplished only twice. HME is easily treated with doxycycline; delayed therapy increases the risk of severe disease and *E. chaffeensis* is not susceptible to chloramphenicol in vitro.

Since 1990, at least 43 patients in Wisconsin, Connecticut, New York, Maryland, Florida and Arkansas have been infected with an as yet unnamed *Ehrlichia* species that causes HGE. Infected *Ixodes scapularis* have been found in regions where this disease occurs. HGE is clinically similar to HME, and usually presents as an undifferentiated fever without rash. Leukopenia, thrombocytopenia and mildly elevated liver function tests are frequent. Elderly patients are more likely to have severe disease. Half of the diagnosed patients have been hospitalized, with 9% admitted to intensive care and approximately 5% dying. Cultivation of the causative agent has not yet been achieved, and seroconversion does not occur until convalescence. Serologic tests for HME do not cross-react with tests for HGE, although peripheral blood smears reveal intraneutrophilic morulae in many patients. Therapy with doxycycline results in defervescence within 48 hours. Also, recent reports indicate that LD patients with prolonged illness that is unresponsive to antibiotics, especially amoxicillin,

may have concurrent infections with *Ehrlichia sp.* Florida added ehrlichiosis to its list of notifiable diseases in 1996.

In Florida, 23 cases of ehrlichiosis were reported between 2000-2002, of which 16 do not have a travel history outside of the state. Most Florida-acquired cases reported exposure in northeastern or panhandle counties.

Chapter 2

Arthropod-borne Disease Control Coordination

Control of arthropod-borne diseases in Florida is coordinated through interagency cooperation at the state and local levels. Intensification of surveillance and initiation of control measures occur in response to evidence of increased transmission in nature. Different agencies become involved at various times during routine surveillance. Therefore, a crucial part of a good surveillance program is to disseminate information to the proper agencies and persons.

Roles and responsibilities

I. Department of Health (DOH) County Health Department (CHD) Activities

Contact: local county health departments

- Conduct epidemiologic investigation to search for new, undetected cases and classify cases as to time (chronological distribution of cases), place (geographic distribution of residence and place of likely exposure) and person (demographics of cases).
- Facilitate submission of diagnostic specimens from physicians and hospitals as required.
- Collect reports of suspected, probable, and confirmed human cases of SLE, WN, EEE, LD, RMSF and ehrlichia. Confirmed and probable cases are reportable under Chapter 381, Florida Statutes.
- Participate in appropriate sentinel avian and horse surveillance activities.
- Communicate with the appropriate mosquito control personnel, school boards, media and public, etc. and coordinate plans for prevention and control activities.
- Provide community information and education as required.
- Coordinate with the DOH Bureau of Community Environmental Health and with mosquito control to issue health alerts to the media or to the public.
- Report human cases in Merlin.

II. DOH Bureau of Laboratory Services Activities

Contact: Department of Health Laboratory, Tampa, (813) 974-8000; Department of Health Laboratory, (904) 791-1540.

- Conduct appropriate tests for detection of arthropod-borne diseases in human, equine and avian surveillance samples.
- Report by telephone the results of all probable and confirmed human serologic or virologic tests to the CHD, the Bureau of Community Environmental Health, and to the attending physician. Follow-up written reports are submitted as soon as possible.
- Report by fax the results of all virus infected birds to the Bureau of Community Environmental Health and the CHD.
- Prepare weekly summary reports indicating the number of sentinel sera submitted, number tested, and number positive by county.

III. DOH Division of Environmental Health, Bureau of Community Environmental Health Activities

Contact: Bureau of Community Environmental Health, (850) 245-4299.

- Direct statewide surveillance, prevention and control programs for human arthropod-borne diseases.
- Provide guidelines for sentinel SLE and WN virus surveillance.
- Conduct epidemiologic analyses of data from CHDs and laboratories.
- Conduct or participate in epidemiologic investigations.
- Distribute epidemiologic reports to CHDs, mosquito control agencies, physicians and veterinarians, CDC and other interested parties.
- Maintain information connectivity among agencies via appropriate medium including weekly electronic *EpiUpdate*, website development and as needed SLE conference calls.
- Recommend health alerts to the State Health Officer.
- Conduct active EEE and WN case surveillance program with Florida veterinarians.
- Coordinate prevention and control activities with DACS, DEP, Florida Tourism Board, mosquito control agencies and other key organizations.
- Coordinate with CDC in interstate and national research, prevention and control efforts.

IV. DOH State Health Office

Contact: Public Information Office, (850) 245-4111

- Reviews press releases as appropriate.
- Issue medical alerts.
- Coordinate media response to medical alerts.

V. Department of Agriculture and Consumer Services (DACs) Bureau of Entomology and Pest Control Activities

Contact: Bureau of Entomology and Pest Control, (850) 922-7011.

- Coordinate with the Bureau of Community Environmental Health and with local county health departments before releasing vector data to the media or to the public.
- Provide technical support, mosquito control and other services as needed to local mosquito control programs and CHDs.
- Facilitate the sharing of mosquito control personnel and equipment between districts, as allowed for in Florida Statutes 388.231 and 388.351.

VI. DACS Division of Animal Industry and Bureau of Diagnostic Laboratory Activities

Contact: State Agriculture Veterinarian, (850) 410-0900; State Diagnostic Laboratory (veterinary), (407) 846-5200.

- Direct statewide surveillance for animal arthropod-borne diseases.
- Conduct appropriate tests for detection of arthropod-borne diseases in animals.
- Report findings to the DOH Bureau of Community Environmental Health on a regular basis.

VII. Mosquito Control Agencies

Contact: local mosquito control agencies or the Florida Coordinating Council on Mosquito Control at (850) 922-7011.

- Conduct appropriate mosquito and arbovirus surveillance as feasible.

- Provide larvicide and adulticide applications as appropriate and feasible.
- Provide adequate avian serosurveillance of most likely sites of SLE and WN virus activity (maintain and monitor flocks and collect and process blood samples) as feasible.

VIII. Florida Universities

Contact: FMEL, (772) 778-7200; PHEREC, (850) 872-4184.

- Provide arthropod-borne disease research at: the Florida Medical Entomological Laboratory (FMEL), University of Florida; the John A. Mulrennan, Sr. Public Health Entomology Research and Education Center (PHEREC) as well as Florida A&M University and University of South Florida.
- Distribute research findings.
- Provide consultation and technical assistance to disease and arthropod control agencies.

IX. Department of Environmental Protection

Contact: Fisheries Management, (850) 922-4340.

- Coordinate efforts for intensified mosquito spraying in protected wetlands as needed during health alerts.
- Provide consultation and technical assistance as required.

X. Florida Tourism Marketing Corporation

Contact: Visit Florida USA, (850) 488-5607.

- Provide timely and accurate arboviral prevention information to attractions, hotels/motels and travel agencies.
- Maintain a toll-free number, 888-735-2872, with appropriate health information for people wishing to visit the state.

XI. Physician/Hospital Activities

Contact: local physicians and hospitals or the Florida Medical Association at (850) 224-6496.

- Report suspected cases of arthropod-borne diseases to the CHD as required by law.
- Submit appropriately timed specimens for confirmation of clinical diagnosis (e.g., CSF and sera, or paired sera drawn at least 1 week apart).

XII. Veterinarians

Contact: local veterinarians or the Florida Veterinary Medical Association at (407) 851-3862.

- Report suspected cases of EEE, WN, LD and ehrlichiosis to the State Veterinarian and the CHD as required by law.

XIII. Centers for Disease Control and Prevention, Division of Vector-Borne Infectious Diseases

Contact: Division of Vector-borne Diseases, (970) 221-6400.

- Provide technical assistance and laboratory support as required.
- Coordinate with the World Health Organization and its regional offices (e.g., Pan American Health Organization) on international research, prevention and control.

Notification and Public Information of arboviral surveillance results

On a weekly basis, DOH will summarize the surveillance data and email the information to the Interagency representatives. DOH will also provide this information to:

The DOH County Health Departments (CHDs)
Centers for Disease Control and Prevention

The interagency partners will distribute the information as follows:

- DACS Bureau of Entomology and Pest Control (BoEPC) will notify:
All organized Mosquito Control Districts
- DACS Division of Animal Industry (DAI) will notify:
Animal Industry Organizations
Veterinarians
- FWCC and Department of Environmental Protection (DEP) will notify:
Regional biologists
Wildlife rehabilitators
- The DOH Bureau of Laboratory will notify:
Sample submitter as results become available

The DOH will be responsible for release of public information regarding recommended public precautions. Local organized mosquito control districts, with the assistance of BoEPC, will be responsible for release of public information regarding mosquito control activities. BoEPC will be responsible for release of public information regarding mosquito control activities in those regions of the state where there is no local organized mosquito control units. DAI will be responsible for release of public information regarding animal health issues.

For the purposes of coordinated local responses and possible intensification of integrated vector control, county health department (CHD) epidemiologists should share nonidentifying case locality and onset information of human arbovirus cases under investigation with local mosquito control districts. DOH will notify the workgroup members by email of the county of residence of such suspect cases.

The interagency partners will strive to immediately share significant new information with each other and the other individuals and organizations listed in this section in order to assure the most rapid response possible to new developments.

Chapter 3

Arthropod-borne Disease Monitoring Activities

The ideal arboviral surveillance program measures the amount of viral amplification and transmission in nature and reliably provides information on the risk of human disease. A complete surveillance program consists of monitoring arboviral seroconversion rates in sentinel chickens, weather patterns, the abundance of vector and amplification host species, and the incidence of human and animal disease. The ultimate goal of surveillance is to increase our ability to predict when and where arboviral transmission to humans is likely to occur so that vector and disease control activities can be implemented prior to the beginning of an epidemic. Continuous local surveillance is also invaluable in monitoring both the progress and the cessation of periods of epidemic risk to man.

I. Sentinel Chicken Serology and Mosquito-Borne Viruses

Mosquito-borne arboviruses are found in mosquitoes throughout Florida during most of the year. Sentinel chickens can be infected with mosquito-borne viruses via the bite of an infected mosquito during any month, but transmission is most often reported between August and November.

Sentinel chickens are frequently infected with SLE and WN viruses but reports of sentinels with EEE virus infections are less common. This is likely due to the extremely focal distribution of EEE virus in Florida and the low probability that sentinel flocks are located in EEE virus transmission zones. Therefore, sentinel chicken surveillance may be less useful for predicting EEE transmission to humans. However, during years of heavy EEE transmission in Florida, including 1978, 1991, and 1997, EEE virus transmission was reported in sentinel chickens over a wide area indicating a generalized risk of EEE virus transmission to humans throughout the traditional Florida EEE virus transmission zone.

Local health and mosquito control agencies should use sentinel chicken flocks to assess local mosquito transmission of WN and other arboviruses. Local governments without mosquito control and/or sentinel chicken surveillance capabilities should work to establish programs in uncovered areas. Testing of sentinel chicken sera for virus and/or antibody will be conducted by the DOH Tampa laboratory and results reported to submitters and participating programs as quickly as possible.

Sentinel chicken programs are maintained by mosquito control districts and/ or county health departments, depending on local resources and priorities. Such programs entail determining flock placement; flock care; weekly collection, processing, and shipping of blood specimens; and notification of appropriate agencies and persons regarding seroconversion data. Under certain conditions, “backyard” juvenile (birds hatched during the sample year) chickens (i.e., birds maintained for other purposes) can be monitored.

Under ideal circumstances, sentinel chicken flocks should be located in every Florida county because mosquito-borne arbovirus transmission can be quite focal and spread rapidly. When flocks are not maintained in a county, that CHD often relies on the results of sentinel chicken surveillance in contiguous counties to aid in decision-making. ***Because of the introduction of WN virus into Florida in 2001 chicken surveillance should be conducted year-round throughout the state.***

Note: Chickens are not known to transmit mosquito-borne viruses directly to people.

A. Sentinel Chicken Flock Information

- The surveillance site should be permanently located in an area free from public access and vandalism. Mosquito control personnel should be consulted for advice on flock placement in counties where CHDs maintain flocks.

- The location of each flock (i.e., maps and GPS coordinates) should be reported to the Bureau of Community Environmental Health each January. The reporting form can be found in Appendix D.
- The number of flocks maintained in each county depends on the size of the county and the resources available for maintaining a sentinel chicken surveillance program. However, a minimum of six chickens per flock is suggested to maintain uninterrupted arboviral surveillance around the vicinity of the flock.
- Sentinel flocks should be located in a variety of habitats throughout the county. These should include, but are not limited to, hardwood hammocks, pine flatwoods, coastal habitats, freshwater marshes, saltwater marshes, residential areas, city and county parks, and urban centers.
- Backyard chicken flocks selected for retrospective surveys should be located within two to three miles of mosquito breeding areas. During a medical alert, chicken flocks within a two-mile radius of a human case may be sampled.
- Female Leghorn, Barred Rock, Rhode Island Red or Minorcan chickens that reach the age of 10-12 weeks before being placed in the field are ideal for surveillance (game chickens are not recommended). All-hen flocks may be preferred in some urban areas when cocks crowing might annoy residents.
- The local county agricultural extension agent can be contacted to obtain information for contacting local chicken breeders. If a local source of chickens is not available, assistance may be obtained from neighboring counties or mosquito control personnel.
- Each chicken must be properly identified by a uniquely numbered wing or leg band-(e.g. available from National Band and Tag Company at 859-261-2035).
- Animal care workers should take precautionary measures when handling chickens and when conducting routine maintenance of cages. Workers should wear latex gloves to protect against contact with potentially infected chicken feces. Chicken feces should be treated carefully and properly disinfected and disposed.

B. Husbandry

- Housing should be constructed in such a manner that the chickens can be protected from the elements (shade and protection from rain is required) and from predators. It is recommended that cages be maintained above the ground.
- A raccoon/fox-proof wired (or double wiring) coop with a strong door and a secure lock to the entrance used for feeding and bleeding purposes should be sufficient to protect the chickens. Mosquitoes must have free access to the coop interior.
- Housing should be adapted to the condition of the terrain and should have adequate slope to keep the ground dry.
- Chickens should be fed in accordance with feed manufacturer's recommendations, including the addition of chicken scratch. Sufficient amounts of fresh water should be supplied to the flocks and cages should be cleaned on a regular basis.
- A separate flock of chickens should be kept in a mosquito-proof building, to replace chickens lost due to seroconversion or mortality.

C. Bleeding Schedules/Record Keeping

- Accurate records should be maintained for future reference with detailed information on the location of the site (exact address and GPS coordinates), surrounding vegetation, and weather conditions during the surveillance season.
- **All chickens in the flock should be bled every week.**
- Arboviral positive individuals are confirmed by a second Hemagglutination Inhibition (HI) positive blood specimen (EEE) or by IgM Elisa and/or Plaque Reduction Neutralization Tests (WN, SLE). Antibody

positive chickens may revert to false HI negative status on later serum samples, thus, chickens that are reported as confirmed positive should be removed from the flock and replaced with a baseline negative bird from the holding flock.

- The weekly seroconversion rate is the number of arboviral positive chickens divided by the number of birds tested. Seroconversion rates can be calculated for the state, county, or individual flocks.
- Serologically negative chickens may be bled throughout the season, but all chickens should be replaced annually with new birds early in the year (April-May).
- **Chickens that seroconvert or die should be replaced with a non-immune chicken having a NEW band number.** Notify the laboratory as to dead/missing chickens and their replacements.

D. Instructions for Bleeding Chickens

A blood collection kit" should be assembled for use in the field. A plastic craft tray or small, light tool box should contain: needles, syringes, serum separator tubes, latex gloves, two pencils or sharpie markers, a small tightly closed plastic container of alcohol-soaked cotton balls, a checklist of chicken wingband numbers by site, insect repellent and waterless hand disinfectant/cleaner for the worker. In addition, bring a sharps container, appropriate disposal bags for waste, and a small cooler of ice (ice is useful for hemostasis if gentle pressure fails to assist with clotting).

Bleeding should be undertaken only by appropriately trained professionals. A person working alone may bleed chickens (a chicken restrainer to facilitate this is described in *Mosquito News* 3(2):357-359, 1986). Two field personnel can make the process easier. Once securely restrained, the bird should be placed on its side and the opposite wing extended for easiest access to the vein that is to be bled:

- A Personnel must take appropriate personal precautions to prevent contact with potentially infectious blood. Personal protection should include, at a minimum, latex gloves.
- B Stretch out a wing to expose its underside. Alternate wings each time the chicken is bled in order to allow healing. (Some may choose to take samples from jugular veins).
- C Pluck feathers where the wing joins the body to expose the vein. Wet the area with alcohol to make the vein more readily visible and to clean the venipuncture site.
- D Carefully insert into the vein, bevel side up, a 23 or 25-gauge 0.5-inch needle (depending on the size of the vein) fitted to a 3cc syringe. Use a new needle and syringe for each chicken.
- E Withdraw 1.5 to 2.0cc of whole blood by drawing on the plunger *slowly* in order to keep the vein from collapsing.
- F Remove needle and apply gentle pressure with alcohol-soaked cotton ball at the site of venipuncture for hemostasis.

Note: Latex gloves should be worn during the entire bleeding procedure. Hands should be cleaned with an alcohol based disinfectant after removing gloves and the gloves disposed of properly as biohazardous waste. Hand sanitizers containing around 70% alcohol are most effective. If the phlebotomist is stuck by a needle during the bleeding procedure, the chicken blood needs to be tested for virus. Contact the Tampa Laboratory at 813-974-5990 for shipping directions.

- G Dispense the blood slowly into a 4-inch commercial serum separator tube. (Tubes can be purchased from Fisher Scientific, 1-800-766-7000.) To reduce hemolysis, uncork the tube, carefully recap and remove the needle from the syringe and slowly express the blood into the tube. The use of these tubes precludes the need to transfer serum and label to a second sterile tube, thus reducing the chance of mislabeling a specimen, and saving technician time. The use of such tubes reduces the rate of bacterial contamination and produces more useable serum.
- H Label each vial using a waterproof marking pen or pencil with the following information:
-correct bird number from the permanent wing tag or leg band -- **important!**

-flock site location

-collection date

- I Lay tubes on their side (this increases serum yield). Keeping tubes on wetpacks helps reduce hemolysis (rupturing of red blood cells).
- J If possible, centrifuge for 15 minutes at 1200rpm, trapping the clot in the bottom of the tube.
- K The tube may be shipped directly to the Bureau of Laboratories, Tampa without decanting the serum. Contact the Tampa Laboratory for shipping containers. See Appendix D for a copy of the laboratory form.

NOTE: All needles should be disposed of in appropriate "sharps" containers and disposed of properly as biohazardous waste. All containers etc. that have come in contact with blood should also be disposed of properly. Disinfection could be accomplished by heat sterilization or chemical sterilization in disinfectant.

- K. **Include a completed "Chicken Arbovirus Surveillance Serology" sheet with serum shipped to the Bureau of Laboratories.** Samples received before noon on Wednesday will have HI test results reported on the following Friday.

E. Serum Testing/Data Dissemination

Sentinel chicken sera are tested at DOH Bureau of Laboratories, Tampa (contact the laboratory at 813-974-8000 or SC 574-8000).

The Tampa lab communicates the results weekly to the county coordinator submitting specimens as well as the county health department, the DOH Bureau of Community Environmental Health and the DACS Bureau of Entomology and Pest Control.

II. Dead Bird Reporting and Testing

The WN virus found in the United States caused morbidity and mortality in many bird species. In some species, especially crows, there has been substantial mortality due to WN infection to detect the presence of the virus in a geographic area. Thus, monitoring dead birds is considered a tool for WN virus surveillance. In Florida, bird mortality sightings from various agencies and the public are to be reported to www.wildflorida.org/bird/. If the dead bird carcass is in the appropriate condition for WN virus diagnostic testing, the carcass and a laboratory form will be submitted by DOH Environmental Health, DACS, FWCC, mosquito control staff, veterinarians or wildlife rehabilitators to the DOH Tampa laboratory to be necropsied and tested using PCR assay and/or virus isolation. The laboratory submission form can be found in Appendix D. This testing should take less than 2 weeks.

III. Surveillance of Human Disease

SLE, WN, EEE, LD, RMSF and ehrlichia are reportable human diseases in Florida. County health departments provide case information to the Bureau of Community Environmental Health for data analysis and dissemination. The surveillance case definitions for these diseases are outlined in Appendix E.

IV. Laboratory Testing Protocol

At the DOH Laboratories, sera collected from sentinel chicken flocks, wild bird samples and clinical human cases are tested for EEE, SLE and WN virus with 3 different serological assays according to the following algorithm: All specimens are screened using an HI assay to detect alphavirus (EEE virus), and/or flavivirus (SLE or WN virus antibodies). Sentinel chicken sera that are flavivirus positive are tested in a WN virus IgM ELISA assay. A repeat serum is tested on WN virus IgM negative chickens. WN virus IgM equivocal

sera may be assayed by serum neutralization for confirmation of etiology. HI flavivirus antibody positive wild bird or mammalian sera are assayed by serum neutralization to confirm the etiological agent. HI alphavirus positive chickens are confirmed by HI testing of a second serum specimen. For interpretation of human test results see Appendix F.

V. Equine surveillance

Equine surveillance is also used to assess the impact of WN and EEE in the state. Veterinarians should send equine sera to the DACS laboratory for evaluation. Results should be available within a week.

VI. Weather analysis – Rainfall Monitoring

Daily rainfall and groundwater accumulations are important meteorological factors to track when attempting to predict changes in vector abundance as well as viral amplification and transmission. Monitoring daily rainfall is important for three reasons. First, the length of the south Florida dry season is an important factor in determining the potential survival of overwintering and potentially infected *Cx. nigripalpus* mosquitoes. During years with a long, dry season (i.e., January through June), there is a lower potential for virus transmission during the following autumn. If the dry season is short, as in 1990, viral amplification and transmission can begin as early as May or June. Second, once the dry season ends, heavy spring rains allow a quick, early season buildup of vector mosquitoes. Finally, daily rainfall patterns are responsible for driving the overall behavior of this mosquito species by determining when and where eggs are laid, when host seeking and biting occurs, and when the virus is transmitted.

Rainfall data is available from the National Weather Service. For more localized information, however, it is often necessary to use independent measurements. To monitor daily rainfall, fence post style rain gauges are read, emptied, and the amount of rainfall recorded at roughly the same time each day. Annual rainfall records include the timing, amount, and intensity of rain at the beginning of the wet season. This alerts personnel to a potential buildup of the vector population. Daily rainfall records throughout the wet season may show patterns of heavy rain (> 2 inches) followed by 10- to 14-day droughts. These conditions are ideal for allowing extrinsic incubation of the virus in infected vectors and for synchronizing vector egg laying, blood feeding and potential virus transmission. Finally, it is important to know when the dry season begins, as this may mark the end of virus transmission for that year.

VII. Mosquito Monitoring

The accurate measurement of vector abundance and population structure is a critical component of arboviral surveillance. Factors such as vector movement, blood feeding, egg laying and the age of the population determine whether there is a high or low risk of viral transmission and the potential for human infection. The number of mosquitoes collected is not as important as the day-to-day changes in the number collected. Therefore it is the quality of collections, not the quantity, which is important. Ideally, the method of surveillance and sampling sites should remain constant from year-to-year, allowing comparison between years.

A. Trapping Mosquitoes

Current methodologies for trapping mosquitoes are available from the Florida Coordinating Council on Mosquito Control or local mosquito control agencies. Printed or diskette copies of Florida Mosquito Control: The State of the Mission as defined by mosquito controllers, regulators, and environmental managers, are available from the Florida Medical Entomology Laboratory, University of Florida/IFAS, 200 9th Street SE, Vero Beach, Florida 32962, (772) 778-7200, or downloaded from FMEL web page: <http://www.ifas.ufl.edu/~veroweb/wpaper.htm>.

Once collections are counted, the number of mosquitoes in each group for each species should be entered into a database for graphical presentation or plotted manually so that day-to-day changes in mosquito abundance can be readily seen. Age determinations allow for identification of periods in which the risk of viral transmission is highest.

B. Viral Assay in Mosquitoes

There is no history of prospective arbovirus surveillance in Florida involving evaluation of SLE, EEE or WN virus infection rates in mosquitoes. It is clear that during epidemic periods, high SLE virus infection rates can be demonstrated in *Culex nigripalpus* mosquitoes.

If implemented, surveillance based on viral assay of mosquitoes would require several years of operation to evaluate its sensitivity and specificity for detecting periods of elevated risk of arbovirus transmission. Surveillance of mosquito infections should not supplant other sources of information pertinent to arbovirus activity (e.g., transmission to sentinel and/or wild vertebrates, real-time monitoring of local *Cx. nigripalpus* population dynamics, rainfall data).

Each organization performing mosquito viral assays should provide test results to the Department of Health Arbovirus Surveillance Coordinator for inclusion in the statewide database. This should include assay method for positive pools, number of pools and number of individuals per pool, species, date and site collected and agent detected. For negative pools, number of pools of each species should be provided.

It is essential that laboratories conducting viral surveillance with mosquitoes (including VecTests) provide appropriate safe procedures for working with BL-2, BL-3 pathogens. Minimal requirements include latex gloves, eye-face protection, disinfection of all materials and supplies and protection against potential aerosols generated during homogenization procedures. For guidelines on trapping and testing mosquitoes for WN virus see Appendix G. The laboratory submission form for mosquito testing can be found in Appendix D.

VIII. Tick Monitoring

Diagnosis of LD, RMSF, and ehrlichiosis cannot be accurately or reliably accomplished through tick identification or by examining ticks for the presence of the disease agents. However, tick collections may be helpful in determining vector species and foci of infection, but only after tick-borne disease has been medically confirmed. Tick surveys are advisable in counties where tick-borne diseases are known to be endemic and when sufficient information exists concerning a specific locality where transmission has occurred. Technical assistance in conducting such surveys may be arranged by contacting the PHEREC, phone (850) 872-4184.

Four tick species are suspected as potential vectors of LD in the southeastern U.S.: *Ixodes scapularis* (the black-legged tick), *Amblyomma americanum* (Lone star tick), *Amblyomma maculatum* (Gulf Coast tick) and *Dermacentor variabilis* (American dog tick). None have been adequately incriminated as the primary vector, though the black-legged tick is the most likely vector of LD in the southeast. This is because it has exhibited a greater capability of transmitting *Borrelia burgdorferi* under laboratory conditions and has been more commonly found naturally infected in the field. Important tick vectors in the southeast for RMSF include *D. variabilis* and *A. americanum*. The most likely tick vector for human monocytic ehrlichiosis is *A. americanum*; for human granulocytic ehrlichiosis *I. scapularis*.

All of these ticks require three different hosts to complete a life cycle consisting of egg, larval, nymphal and adult stages. After hatching from eggs deposited on the ground usually in grassy, brushy or wooded areas, tiny six-legged larval ticks (also known as "seed" ticks) climb on vegetation and wait to cling upon passing hosts. Small rodents (woodland mice), ground birds and reptiles (lizards and snakes) most commonly serve as hosts for larval and nymphal ticks. After obtaining blood meals, larval ticks drop to the ground, molt (i.e., shed their "skin") and develop to eight-legged nymphs. Nymphs follow a similar sequence feeding on a

different host before molting to the adult stage. Adult ticks usually seek larger hosts such as deer, cattle and possibly humans. Under field conditions, each of these species require 1-2 years to complete their life cycle. This period may span, for some, over 3 calendar years for eggs deposited late in the season.

Based on submissions for tick identification to the then HRS Entomology Services office (currently Florida Department of Agriculture and Consumer Services, Bureau of Entomology and Pest Control), the Lone star tick and the Gulf coast tick are the most common human-biting species in Florida.

Chapter 4

Control Measures and Surveillance Response

I. Personal Protection

Education messages should be targeted to at-risk populations (e.g., emphasize high risk of SLE and WN for the elderly) in low-literacy forms and in languages appropriate to the local population. Media should be used, including radio, newspaper, and television public service announcements (see Appendix K).

A. Mosquito-borne Diseases

The effectiveness of public education as a control measure for SLE was demonstrated in the 1997 outbreak. A study of the outbreak by the DOH Bureau of Epidemiology showed that people who had received public health messages were significantly more likely to reduce their exposure to mosquitoes than those who had not heard the messages.

People can protect themselves from mosquito bites (and therefore arboviruses) by using proper window screens, protective clothing and insect repellent. The principal vector of SLE, *Cx nigripalpus*, blood feeds from dusk through dawn with activity most intense at dusk and dawn. Consequently, in an actual or potential epidemic situation, people should be encouraged to avoid mosquito contact at those times of day. The ordinary window screen with 16x16 or 14x18 meshes to the inch will keep out most mosquitoes, including arbovirus vectors. Frequently, mosquitoes follow people into buildings or enter on the host. For this reason, screen doors should open outward and have automatic closing devices. Residual insecticide applications on and around screen doors give added protection.

Long-sleeved clothing of tight-woven material offers considerable protection against mosquito bites. Sleeves and collars can be kept buttoned and trousers tucked in socks when mosquitoes are biting. This type of protection may be necessary for people who must work in areas where infected vector mosquitoes are particularly abundant. The use of mosquito netting to protect infants in their cribs may also be indicated in high-risk circumstances.

Applying insect repellent to the skin and clothing may offer relief from mosquito attack. When the potential exists for exposure to mosquitoes, repellents containing DEET (N,N-diethyl-meta-toluamide, or N,N-diethyl-3-methylbenzamide) are recommended. Products with concentrations up to 30% DEET are generally recommended for most situations. Repellents are available as liquids in bottles, in pressurized spray cans and in stick form. When applied to the neck, face, hands and arms, liquid repellent will prevent mosquito bites for two hours or more, depending on the person, the species of mosquito attacking and the abundance of mosquitoes. These repellents can also be sprayed on clothes (DEET will not affect nylon). Many repellents are solvents of paints, varnishes and plastics (including watch crystals, rayon fabrics and fountain pens). Care should be taken not to apply repellents to the eyes, lips or mucous membranes. If additional protection is necessary, apply a permethrin repellent directly to your clothing. Always read the manufacturer's directions carefully before you put on a repellent. Adults should apply repellent to young children. It is not recommended to use DEET on children less than 2 months old. Instead, infants should be kept indoors or mosquito netting used over carriers when mosquitoes are present. (For information about DEET, see Appendix K).

Pressurized aerosol insecticide dispensers can be used in the home to kill adult mosquitoes. Insecticide label directions must be followed. Most of these contain pyrethrin or allethrin. These insecticides have low human toxicity and cause a quick knockdown of mosquitoes. These aerosol dispensers may also

contain a synergist such as piperonyl butoxide and another insecticide, such as diazinon, to kill the insects. Release of the aerosol for a few seconds usually kills most insects in an ordinary-sized room, tent or trailer.

B. Tick-borne Disease

Prevention is the best way to avoid diseases vectored by ticks. Persons involved in outdoor activities in tall grass, brushy or treed areas should follow these instructions:

1. Tuck trouser legs into boots or socks.
2. Use repellents containing up to 30% N,N -diethyl-m-toluamide (DEET, e.g., Off[®], Cutters[®]) and/or the clothing-applied insecticide, such as permethrin (e.g., Permanone[®] Tick Repellent) according to labeled directions.
3. Check to remove crawling ticks at least every three hours while outdoors. Wearing light-colored clothing will make spotting ticks easier.
4. Before going to sleep or after returning indoors, remove and wash clothing or place in a tightly sealed bag for storage until washing. Conduct a full-body check for ticks followed by a shower or bath.
5. Outdoor pets should be checked frequently and treated with an acaricidal shampoo according to labeled directions.

II. Arbovirus Medical Alert

A medical alert is a declaration by the State Health Officer that “a threat to the public health exists” as per Florida Statutes, 388.45. Residents and visitors to the affected area are urged to take personal protective measures against biting arthropods. This official declaration also allows DACS to respond with actions allowing more liberal use of arthropod control measures on certain public lands and movement of mosquito control personnel and equipment into affected counties from other areas of the state as appropriate. The need for a medical alert is determined by the CHD director/administrator after consultation with the State Health Office. Increased sentinel chicken seroconversion rates to SLE, WN or EEE viruses, weather information, vector surveillance (including mosquito trapping), historic arbovirus distribution records, arbovirus cases within domestic and wild animal populations, and the presence of human cases in the same or contiguous counties are important factors to consider when initiating a medical alert.

A medical alert may be initiated in any specific geographic area of the state where there has been:

1. A confirmed human case of SLE, WN, EEE, or other arbovirus of comparable significance thought to have been contracted in Florida
OR
2. Substantial increases over long-term county norms in the number of seroconverting sentinel chickens, an increase in the proportion of flocks containing seroconverting sentinels, or an unusual timing of multiple seroconverting sentinels
OR
3. A rapid increase in the SLE, WN or EEE seroconversion rate in a single sentinel flock over a two-week period
OR
4. More than one mosquito pool with all of the following characteristics: infected with a virus (SLE, WN, or EEE), capable of infecting humans, collected within 5 miles and 10 days of another infected mosquito pool, causing a significant increase in the rate of mosquito infection when compared with the usual rate for the county
OR
5. A cluster of equine or ratite (e.g., ostriches, emus) WN or EEE cases (e.g., two or more cases within five miles and ten days of each other).

The CHD Director/Administrator facilitates arbovirus medical alert activities. This includes working closely with the Bureau of Community Environmental Health, mosquito control personnel, physicians, veterinarians, emergency rooms and neighboring counties. DACS Bureau of Entomology and Pest Control provides technical support and leadership to CHDs and county mosquito control programs as needed during a medical alert. SLE or WN epidemic activity may remain localized to a city or county; however, Florida's last two SLE and WN outbreaks were more widespread, with multiple counties affected. Further, SLE, WN or EEE viral activity can have "hot spots" or discrete transmission foci interspersed within areas of little or no virus transmission. Therefore, epidemic and medical alerts and control measures cannot always be uniformly applied in all areas of the state.

- The CHD in the affected county will notify:
 1. Community health care providers concerning the potential for transmission of SLE, WN or EEE virus to people, and the need for physicians and veterinarians to report new cases.
 2. The County Mosquito Control Director.
 3. CHD Directors/Administrators and Mosquito Control Directors in contiguous counties of the medical alert.
 4. Local media, education representatives, senior citizen groups and other citizen groups as appropriate.
- The Bureau of Community Environmental Health will notify DACS and DEP within 24 hours of the declaration of a medical alert (Florida Statutes 388.45).

A. Intensified Public Education

The goals of public education are to inform the public about personal protection measures (described above), provide information and prevent panic. CHDs in coordination with the county mosquito control programs may:

1. Issue advisories to minimize outside evening and early morning activities for citizens of affected counties (e.g., activities such as camping, evening and nighttime fishing, etc. are ill advised).
2. Advise persons who do continue to spend time out-of-doors in the evening, nighttime or early morning hours to wear protective clothing (long-sleeved shirts, long pants) and to use insect repellent.

Remember the 5 D's for arbovirus prevention:

- Dusk and Dawn (Avoid being outdoors when mosquitoes are most active.),
 - Dress (Cover your skin with clothing.),
 - DEET (Use mosquito repellent on bare skin and clothing.) and
 - Drain (Remove standing water in which mosquitoes can lay their eggs.)
3. Educate the public about the nature of the public health threat that exists and the level of risk involved (including age-specific risk).
 4. For EEE, attempt to gain immediate control of infected adult mosquito populations by use of insecticides applied by ground or aerial applications, as appropriate. Implementation of intensified larviciding programs to reduce future adult populations and elimination of mosquito breeding areas, where applicable, may also be necessary.

III. Vector Control

A. Reduce Mosquito Breeding Areas

Communities and residents should:

- Eliminate standing water in depressions, barrels, containers and drains.

- Repair leaking septic tanks, cesspools and drainfields.
- Remove old tires.
- Stack containers upside-down so they do not accumulate water.

B. Tick Control

Area pesticide spraying programs for ticks are not practical for many situations. Consultation with PHEREC is advisable before considering this procedure. Deer feeders equipped with self-treating permethrin insecticide dispensers may be useful in reducing ticks in locations with large deer populations.

IV. Arbovirus Coordinated Response Plan

The number of WN virus detections in Florida bird populations, sentinel chickens, and humans is unpredictable, since this virus is new to the Florida ecosystem. The responses to the detection of a WN, EEE or SLE virus positive are based on the information that will allow appropriate risk assessment of transmission to humans, such as: 1) the number of animals detected with the virus in a region, 2) the time period during which the positives were observed, 3) the methods used to make the detections, 4) the season, 5) the abundance of suspected mosquito vectors, 6) the abundance of WN infected vectors, and 7) the mosquito transmission rate. Public interest can be expected to be very high, with concern on the part of public health, mosquito control and the public increasing as the numbers of detections increases, particularly if there are human cases.

The participating Arbovirus Response Plan agencies will provide the following activities based on the potential for human infection in a particular region of the state each year:

Response Level 1: Initial detection of arbovirus nucleic acid or antibody in sentinel flocks, wild or domestic birds, mammal or mosquitoes in a particular region of the state:

- Consider participating in weekly teleconference calls conducted by the DOH
- Maintain surveillance activities that may already be in place in that region or coordinate additional surveillance, i.e., consideration of sentinel chicken flock surveillance to monitor mosquito transmission
- Maintain vector control activities that are already in place in that region
- Solicit dead bird submissions for testing from the public and appropriate local agencies in the region if already not underway
- Consider local advisory to provide public information on personal protective measures
- Provide information to physicians and hospitals on symptomology and appropriate specimen submission

Response Level 2: Wide-spread Detections in sentinel flocks, wild or domestic birds, mammals or mosquitoes

- As above, with consideration of DOH-declared medical alert in affected area.
- Increase levels of surveillance activities – heighten state and local efforts to collect wild birds, vector mosquitoes, and place more sentinel chickens, if possible
- DACS will consider the need to issue a mosquito declaration in those counties needing additional mosquito control measures based on the State Health Officer’s declaration of a threat to the public health or if a

threat to animal health exists. This includes consideration of requesting external resources for use in aerial adulticiding activities. This would be based on a local assessment of vector mosquitoes in the region and the effectiveness of aerial adulticiding in reducing suspect vector populations.

Response Level 3: Detection of 1 or more human cases with Florida-acquired WN virus

- As above, with consideration of DOH-declared medical alert in affected area if not already issued.
- Aerial adulticiding would be considered based on the likely impact on the suspect vectors. This would include requests for assistance using from other agencies with aerial adulticiding capability as appropriate.

Response Level 4: Detection of widespread distribution of human cases in conjunction with a weather-related disaster (e.g., hurricane or flooding event).

- As above, with:
 - A declaration of Medical Alert if not already issued.
 - Implementation of Emergency Operations Center (EOC) Level 2 for Emergency Support Function (ESF) 8 and ESF 17 to coordinate response activities, attachment of representatives of agencies participating in WN virus Coordination Group to these ESFs for duration of Level 2 activation
 - Coordination of requests for vector control assistance through EOC

Coordination of public information dissemination through EOC

Sections 388.231 and 388.351, Florida Statutes, authorize the movement of mosquito control personnel and equipment into affected counties from other counties. This is coordinated through the DACS Bureau of Entomology and Pest Control Office.

The main vector for SLE, *Cx. nigripalpus*, is difficult to control due to the adults' ability to fly several miles and its wide range of larval habitats. The public should be educated about the challenges to controlling this vector.

C. Tick Control

Area pesticide spraying programs for ticks are not practical for many situations. Consultation with PHEREC is advisable before considering this procedure. Deer feeders equipped with self-treating permethrin insecticide dispensers may be useful in reducing ticks in locations with large deer populations.

Acronyms/Definitions

Ae.:	Abbreviation for mosquitoes in the genus <i>Aedes</i>
Arbovirus:	Arthropod-borne virus
Arthropod:	Animals in the phylum which includes insects (mosquitoes, flies, etc.) and arachnids (ticks, spiders, etc.)
CHD:	County health department
CF test:	Complement fixation test
Cq.:	Abbreviation for mosquitoes in the genus <i>Coquillettidia</i>
Cs.:	Abbreviation for mosquitoes in the genus <i>Culiseta</i>
Cx.:	Abbreviation for mosquitoes in the genus <i>Culex</i>
DACS:	Department of Agriculture and Consumer Services
DEN:	Dengue fever
DEP:	Department of Environmental Protection
DOH:	Department of Health
EEE:	Eastern Equine Encephalomyelitis
EIA/ELISA:	Enzyme immunoassay/enzyme-linked immunosorbant assay
EM:	Erythema migrans. EM is defined as a skin lesion that typically begins as a red macule or papule and expands over a period of days to weeks to form a large round lesion, often with partial central clearing
Encephalitis:	Inflammation of the brain
FMEL:	Florida Medical Entomology Laboratory
Hemostasis:	The arrest of bleeding
HGE:	Human granulocytic ehrlichiosis
HI/HAI:	Hemagglutination (and antibody) inhibition test used by the DOH Tampa Branch Laboratory for avian serosurveillance
HME:	Human monocytic ehrlichiosis

IFA: Immunofluorescent antibody test

Ig: Immune globulin or antibody (as in IgM, IgG, IgD, IgA or IgE)

LA/LAT: Latex agglutination test

LD: **Lyme Disease**

MA: Microagglutination test

Morulae: Spherical mass (from the word “mulberry”)

Oc.: Abbreviation for mosquitoes in the genus *Ochlerotatus*

PHEREC: John A. Mulrennan, Sr., Public Health Entomology Research and Education Center (Florida A&M University)

RMSF: Rocky Mountain Spotted Fever

Serum/Sera: The clear liquid separated from blood

SLE: St. Louis Encephalitis

SN Index: Serum neutralization index

Surveillance: Close observation for disease detection

Vector: A carrier which transfers infective agents from one host to another

Venipuncture: Puncture of a vein as for drawing blood

WN: West Nile

Zoonosis: Disease of animals transmissible to people

>=: Greater than or equal to

<=: Less than or equal to

DOH LABORATORY EVALUATION OF ARTHROPOD-BORNE VIRAL DISEASES IN PEOPLE

Introduction

A number of clinical syndromes accompany arboviral infection including fever, rash, myalgia, arthralgia, hemorrhagic fever and encephalitis. Serologic surveys indicate that the ratio of inapparent to apparent infections is sometimes quite high. These viruses usually cause an abortive infection characterized by fever, headache and other benign signs. However, a few individuals will develop a clinical infection that may be severe or fatal.

It is important to confirm a specific agent in instances of a suspected infection. This enables appropriate patient therapy and also permits vector control operations designed to limit transmission to additional susceptible human hosts. Confirmation is dependent upon direct viral detection or serologic examinations such as the hemagglutination-inhibition (HI), complement-fixation (CF), serum-neutralization (SN), enzyme-linked immuno-sorbent assay (ELISA) and fluorescent antibody (FA) tests. Interpretation of each of the tests is dependent upon the time after onset of illness, the patient's previous infection with arthropod-borne viruses and serum cross-reactivity within the antigenic complex. In Florida, previous dengue infection or previous Yellow Fever vaccine are the most common factors that can complicate the interpretation of antibody tests.

Available Laboratory Testing

Virus Isolation -- It is rare to isolate SLE virus from blood or cerebrospinal fluid taken during the acute phase of encephalitis due to rapid completion of the viremic stage prior to onset of illness. SLE and WN viruses can be detected in brain tissue collected at necropsy. EEE and WEE viruses are also usually only isolated from the brain. Dengue virus, however, frequently may be isolated from blood during the first few days after onset of illness.

Serum Neutralization (SN) -- Neutralizing antibody contains both IgG and IgM antibody fractions. SN antibody rises early in the course of infection, and may persist for life after some viral infections, specifically SLE or dengue.

Serum IgM Antibody -- The IgM serum fraction is involved in both the SN and HI reactions, but IgM can be detected independently in either serum or cerebrospinal fluid (CSF) using a capture enzyme immunoassay. The presence of IgM is generally a reliable indicator of recent infection. However, a subset of case patients may have persisting serum IgM antibody to flaviviruses, thus somewhat limiting the value of the assay as a measure of recent infection. Since IgM antibody does not cross the blood-brain barrier, its presence in CSF indicates local antibody synthesis in response to a central nervous system infection and is usually diagnostic.

Serum Hemagglutination-Inhibition Antibody (HI) -- Both the IgG and IgM antibody fractions are responsible for the HI reaction. HI titers can become positive quite early in the course of infection, and a rise in titer is diagnostic of recent infection. Crossreactivity within a virus group (e.g., flaviviruses) is common, and can complicate interpretation of results.

Complement Fixation (CF) Antibody -- Compared to HI antibody, CF antibody is later-appearing, more complex-specific and shorter-lived, and can therefore be useful in diagnosing recent infection.

Specimen Collection

When virus isolation is attempted, blood serum, CSF and tissue samples are placed on dry ice immediately after collection and kept frozen on dry ice while in transit to the laboratory. Fluids are kept in standard airtight tubes, and tissue in an airtight container. When serum is to be examined only for antibody, it can be shipped at ambient temperature (do not freeze) provided it has been collected and handled aseptically. At least 2ml of serum or CSF are required for antibody testing.

Shipping Specimens

Sera are sent immediately to the assigned DOH laboratory for HI testing. Tubes are wrapped individually in paper or placed in cardboard containers having dividers so that tubes do not touch (being sure that the vial will not leak). Such containers are readily available from county health departments.

These containers are placed inside a second shipping container. Sera need not be shipped on ice. However, to retard bacterial growth, sera are stored in a refrigerator or freeze, until shipped. Sera are not shipped on a Thursday or Friday because they may arrive at the DOH laboratory on a weekend.

NOTE: UNSEPARATED, WHOLE BLOOD MUST NOT BE SHIPPED TO THE LABORATORY

To expedite receipt of specimens at the laboratory, overnight or 2-day express shipment is suggested. The following must appear on the shipping label:

DOH Central Laboratory - Viral Center
1217 Pearl Street
Jacksonville, FL 32202
Phone (904) 791-1539, 791-1540

OR

DOH Tampa Branch Laboratory - Virology Unit
3602 Spectrum Boulevard
Tampa, FL 33612
Phone (813) 974-5990.

Appendix C

CONTACTS FOR ESTABLISHING SENTINEL CHICKEN FLOCKS

(Note: Listing does not necessarily denote endorsement. Contact established sentinel sites for more information.)

Chicken Suppliers (White Leghorn or Rhode Island Reds suggested)

- Zephyr Eggs (813) 782-1521
- Hillandale Farms (386) 397-1300
- Ben Mather, University of Florida Extension Service (mather@animal.ufl.edu)

Wing/Leg Bands

- National Band and Tag Company: (859) 261-2035

Serum Separator Tubes

- Fisher Scientific: (800) 766-7000, catalog # 02-65714 (13x75mm)

Chicken Cages, Feeders and Waterers

- Stromberg's: (800) 720-1134
- Plans for building cages provided upon request: 850-245-4299 (Hardrick Gay and Delores Miller are gratefully acknowledged)
- Plans for self-feeder, self-waterer provided upon request: 850-245-4299 (Robert Betts is gratefully acknowledged)

Chicken Restrainer Board

- Plans for restrainer available upon request: 850-488-2905 (Delores Miller is gratefully acknowledged)

Chicken Feed

- Available at local feed store



Bureau of Laboratories
 Tampa Branch Laboratory
 3602 Spectrum Boulevard
 Tampa, FL 33612
 e-mail: Lillian_stark@doh.state.fl.us or Deno_kazanis@doh.state.fl.us

phone: (813) 974-5990
 fax: (813) 974-5776
 suncom: 574-5990
 suncom fax: 574-5776

Arbovirus Surveillance Serology

County _____ page _____ of _____

Contact name _____

Address _____

Phone () _____

Fax: () _____

E-mail: _____

Specimen Collection Data

Sample date	Bird #	Site	New	Species	Sex/Age
1.					
2.					
3.					
4.					
5.					
6.					
7.					
8.					
9.					
10.					
11.					
12.					

For Laboratory use only

Date Received: _____

Date Reported: _____

HAI titer

LAB Number	Flavi*	EEE	Comments

Flavi* = Flavivirus - includes SLE & WN

This form must accompany all serum specimens submitted for serologic examination.

Submitter should fill out left side of form completely. **DO NOT SKIP LINES** when listing collected specimens

If bird is new to the flock or first time bled, place an X in the "New" column. Please **Do not write below this line**



Bureau of Laboratories
 Tampa Branch Laboratory
 3602 Spectrum Boulevard
 Tampa, FL 33612
 e-mail: Lillian_stark@doh.state.fl.us or Deno_kazanis@doh.state.fl.us

phone: (813) 974-5990
 fax: (813) 974-5776
 suncom: 574-5990
 suncom fax: 574-5776

Arbovirus Surveillance: Necropsy and Virus Isolation

County _____ **Reported on** <http://wld.fwc.state.fl.us/bird/>
 ___yes ___no

Contact name _____ **E-mail:** _____

Organization _____ **Phone** () _____
 _____ :

Address _____ **Fax:** () _____

Address _____

City/State/zip _____

For DoH Tampa Laboratory Use Only		
Date Received		
DoH LAB #	Molecular Assay Results	Virus Isolation Result

Specimen Collection Data

Collection date	Bird Mortality Database #	Site/Address of Collection <i>OR</i> GPS Coordinates	Species of bird

Please send birds (only recently dead within the past 24 hours) to:

*Florida Department of Health, Bureau of Laboratories, 3602 Spectrum Blvd.,
 Tampa, FL 33612-9401, Attention: Virology (B)*

2003 Sentinel Chicken Surveillance - Individual Site Information

Contact Information

Name _____ **Phone** _____

Email Address _____

Agency _____

Please confirm the following information regarding your flock(s):

County _____ **Unique ID #** _____

Street Address _____

City _____

Zipcode _____

GPS Coordinates _____

Number of Birds In Your Flock _____ as of _____
date

Comments: _____

Please fax (or email) to Kristen Payne at (850) 922-8473.

Kristen_Payne@doh.state.fl.us

SURVEILLANCE CASE DEFINITIONS FOR ARTHROPOD-BORNE DISEASES IN FLORIDA

Acute Arboviral Disease

reporting code = 06220 Eastern Equine Encephalitis (EEE)
 reporting code = 06230 St. Louis Encephalitis (SLE)
 reporting code = 06620 Venezuelan Equine Encephalitis (VEE)
 reporting code = 06210 Western Equine Encephalitis (WEE)
 reporting code = 06250 California/La Crosse Encephalitis
 reporting code = 06630 West Nile Virus (WNV)
 reporting code = 06631 West Nile Fever
 case report form: Encephalitis Case Report

Clinical description

Arboviral infection may result in a febrile illness of variable severity associated with neurologic symptoms ranging from headache to aseptic meningitis or encephalitis. Symptoms can include headache, confusion or other alteration in sensorium, nausea, and vomiting. Signs may include fever, meningismus, cranial nerve palsies, paresis or paralysis, sensory deficits, altered reflexes, convulsions, abnormal movements, and coma of varying degree.

Laboratory criteria for diagnosis

- Fourfold or greater change in serum antibody titer

OR

- Isolation of virus from or demonstration of viral antigen or genomic sequences in tissue, blood, cerebrospinal fluid (CSF), or other body fluid

OR

- Specific IgM antibody by enzyme immunoassay (EIA) antibody captured in CSF or serum. Serum IgM antibodies alone should be confirmed by demonstration of IgG antibodies by another serologic assay (e.g., neutralization or hemagglutination inhibition [HAI]).

Case classification

Confirmed: a clinically compatible case that is laboratory confirmed

Probable: a clinically compatible case occurring during a period when arboviral transmission is likely, and with the following supportive serology: a stable (\geq twofold change) elevated antibody titer to an arbovirus (e.g., \geq 320 by hemagglutination inhibition, \geq 128 by complement fixation, \geq 256 by immunofluorescence, and \geq 160 by neutralization, or \geq 400 by enzyme immunoassay IgM).

Comment: Arboviral encephalitis cannot be distinguished clinically from other central nervous system (CNS) infections. Acute and convalescent sera from reported and suspect cases should be acquired and sent to the State Laboratory.

A COPY OF LABORATORY TEST RESULTS MUST ACCOMPANY THE CASE REPORT FORM.

CDC-Recommended surveillance case definition for West Nile fever

Clinical Description

A non-specific, self-limited, febrile illness caused by infection with West Nile virus, a mosquito-borne flavivirus. Clinical disease generally occurs 2-6 days (range 2-15 days) following the bite of an infected mosquito. Typical cases are characterized by the acute onset of fever, headache, arthralgias, myalgias, and fatigue. Maculopapular rash and lymphadenopathy generally are observed in less than 20% of cases. Illness typically lasts 2-7 days.

Laboratory criteria for diagnosis

- Fourfold or greater change in West Nile virus-specific serum antibody titer

OR

- Isolation of West Nile virus virus from or demonstration of West Nile viral antigen or genomic sequences in tissue, blood, cerebrospinal fluid (CSF), or other body fluid

OR

- Specific IgM antibody by enzyme immunoassay (EIA) antibody captured in CSF or serum. Serum IgM antibodies alone should be confirmed by demonstration of IgG antibodies by another serologic assay (e.g., neutralization or hemagglutination inhibition [HAI]).

Case classification

Confirmed: a clinically compatible case that is laboratory confirmed

Probable: a clinically compatible case occurring during a period when arboviral transmission is likely, and with the following supportive serology: a stable (\geq twofold change) elevated antibody titer to an arbovirus (≥ 128 and ≥ 160 by neutralization, or ≥ 400 by enzyme immunoassay IgM).

Comment:

Some West Nile fever cases progress to West Nile meningitis or encephalitis. Cases meeting the more restrictive case definition of West Nile encephalitis/ meningitis should be reported as such and only once, using the event code 06631.

A COPY OF LABORATORY TEST RESULTS MUST ACCOMPANY THE CASE REPORT FORM.

Dengue Fever

reporting code = 06100
case report form: CDC 56.31A (10/85)
Dengue Case Investigation

Clinical description

An acute febrile illness characterized by frontal headache, retroocular pain, muscle and joint pain, and rash. The principal vector is the *Aedes aegypti* mosquito and transmission usually occurs in tropical or subtropical areas. Severe manifestations (e.g., dengue hemorrhagic fever and dengue shock syndrome) are rare but may be fatal.

Laboratory criteria for diagnosis

- Isolation of dengue virus from serum and/or autopsy tissue samples

OR

- Demonstration of a fourfold or greater rise or fall in reciprocal IgG or IgM antibody titers to one or more dengue virus antigens in paired serum samples

OR

- Demonstration of dengue virus antigen in autopsy tissue or serum samples by immunohistochemistry or by viral nucleic acid detection

Case classification

Confirmed: a clinically compatible case that is laboratory confirmed

Probable: a clinically compatible case with supportive serologic findings (a reciprocal IgG antibody titer of ≥ 1280 or a positive IgM antibody test on a single acute (late)- or convalescent-phase serum specimen to one or more dengue virus antigens)

Comment

Dengue hemorrhagic fever is defined as an acute febrile illness with minor or major bleeding phenomena, thrombocytopenia ($\leq 100,000/\text{mm}^3$), and evidence of plasma leakage documented by hemoconcentration (hematocrit increased by $\geq 20\%$) or other objective evidence of increased capillary permeability. The definition of dengue shock syndrome follows all of the above criteria for dengue hemorrhagic fever and also includes hypotension or narrow pulse pressure (≤ 20 mm Hg).

Acute and convalescent sera from reported and suspect cases should be acquired and sent to the State Laboratory.

A COPY OF LABORATORY TEST RESULTS MUST ACCOMPANY THE CASE REPORT FORM.

Ehrlichiosis, Human

reporting code = 08381 Human Granulocytic Ehrlichiosis (HGE)
reporting code = 08382 Human Monocytic Ehrlichiosis (HME)
reporting code = 08380 Human Ehrlichiosis, Other
case report form: CDC 55.1 (1/01)

Tick-Borne Rickettsial Disease Case Report

Clinical description

A tickborne febrile illness most commonly characterized by acute onset, accompanied by headache, myalgia, rigors and/or malaise. Clinical laboratory findings may include intracytoplasmic microcolonies (morulae) in leukocytes of peripheral smear, cerebrospinal fluid (CSF), or bone marrow aspirate or biopsy, cytopenias (especially thrombocytopenia and leukopenia), and elevated liver enzymes (especially alanine aminotransferase or aspartate aminotransferase).

Laboratory criteria for diagnosis

- Fourfold or greater change in antibody titer to *Ehrlichia* spp. antigen by immunofluorescence antibody (IFA) test in acute- and convalescent-phase specimens ideally taken ≥ 4 weeks apart. HME diagnosis requires *E. chaffeensis* and HGE currently requires *E. equi* or HGE-agent antigen

OR

- Positive polymerase chain reaction assay
- Intracytoplasmic morulae identified in blood, bone marrow, or CSF leukocytes, **and** an IFA titer $\geq 1:64$

Case classification

Confirmed: a clinically compatible case that is laboratory confirmed

Probable: a clinically compatible case with either a single IFA serologic titer $\geq 1:64$ or intracytoplasmic morulae identified in blood, bone marrow, or CSF leukocytes

Comment:

There are two clinically similar yet serologically distinct forms of ehrlichiosis: a) human granulocytic ehrlichiosis (HGE), caused by infection with an *Ehrlichia equi*-like agent and found primarily in the upper midwest and northeast, and b) human monocytic ehrlichiosis (HME) caused by *Ehrlichia chaffeensis* infection and found primarily in the southeastern quadrant of the United States. Distinct primers are used for the PCR diagnosis of HGE and HME. **Acute and convalescent sera from reported and suspect cases should be acquired on all cases and sent to the State Laboratory.**

A COPY OF LABORATORY TEST RESULTS SHOULD ACCOMPANY THE CASE REPORT FORM.

Lyme Disease

reporting code = 06959
case report form: CDC 52.60 (7/90)
Lyme Disease Case Report Form

MERLIN ELECTRONIC SUBMISSION

Clinical description

A systemic, tickborne disease with protean manifestations, including dermatologic, rheumatologic, neurologic, and cardiac abnormalities. The best clinical marker for the disease is the initial skin lesion (i.e., erythema migrans [EM]) that occurs in 60%–80% of patients.

Laboratory criteria for diagnosis

- Isolation of *Borrelia burgdorferi* from a clinical specimen

OR

- Demonstration of diagnostic IgM or IgG antibodies to *B. burgdorferi* in serum or cerebrospinal fluid (CSF) by EIA or IFA screen followed by demonstration of IgM or IgG antibodies by Western Blot (WB) in specimens taken less than 8 weeks after appearance of EM lesions. [IgG WB should be performed on specimens taken >8 weeks after disease onset – IgM WB in the chronic stage does not aid in the diagnosis of late-stage disease]

Case classification

Confirmed: a) a case with EM that is physician and laboratory (EIA and WB) confirmed or b) a case with one late manifestation (as defined below) that is laboratory (EIA and IgG WB) confirmed

Comments

Definition of terms used in the clinical description and case definition:

- *Erythema Migrans*. For purposes of surveillance, EM is defined as a skin lesion that typically begins as a red macule or papule and expands over a period of days to weeks to form a large round lesion, often with partial central clearing. A single primary lesion must reach ≥ 5 cm in size. Secondary lesions also may occur. Annular erythematous lesions occurring within several hours of a tick bite represent hypersensitivity reactions and do not qualify as EM. For most patients, the expanding EM lesion is accompanied by other acute symptoms, particularly fatigue, fever, headache, mildly stiff neck, arthralgia, or myalgia. These symptoms are typically intermittent. A physician must make the diagnosis of EM.
- *Late Manifestations*. These include any of the following when an alternate explanation is not found:
 1. MUSCULOSKELETAL SYSTEM. Recurrent, brief attacks (weeks or months) of objective joint swelling in one or a few joints, sometimes followed by chronic arthritis in one or a few joints. Manifestations not considered as criteria for diagnosis include chronic progressive arthritis not preceded by brief attacks and chronic symmetrical polyarthritis. Additionally, arthralgia, myalgia, or fibromyalgia syndromes alone are not criteria for musculoskeletal involvement.
 2. NERVOUS SYSTEM. any of the following, alone or in combination: lymphocytic meningitis; cranial neuritis, particularly facial palsy (may be bilateral); radiculoneuropathy; or, rarely, encephalomyelitis. Encephalomyelitis must be confirmed by demonstration of antibody production against *B. burgdorferi* in the CSF, evidenced by a higher titer of antibody in CSF than in serum. Headache, fatigue, paresthesia, or mildly stiff neck alone is not criteria for neurologic involvement.
 3. CARDIOVASCULAR SYSTEM. acute onset of high-grade (2nd° or 3rd°) atrioventricular conduction defects that resolve in days to weeks and are sometimes associated with myocarditis. Palpitations, bradycardia, bundle branch block, or myocarditis alone are not criteria for cardiovascular involvement.
- *Exposure*. Exposure is defined as having been (≤ 30 days before onset of EM) in wooded, brushy, or grassy areas (i.e., potential tick habitats) in a county in which Lyme disease is endemic. A history of tick bite is not required.
- *Disease Endemic to County*. A county in which Lyme disease is endemic is one in which at least two confirmed cases have been previously acquired or in which established populations of a known tick vector are infected with *B. burgdorferi*.

A copy of specific laboratory test results must accompany the case report form.

Malaria

reporting code = 08460
case report form: CDC 54.1 (10/97)
Malaria Case Surveillance Report

Clinical description

Signs and symptoms are variable; however, most patients experience fever. In addition to fever, common associated symptoms include headache, back pain, chills, sweats, myalgia, nausea, vomiting, diarrhea, and cough. Untreated *Plasmodium falciparum* infection can lead to coma, renal failure, pulmonary edema, and death. The diagnosis of malaria should be considered for any person who has these symptoms and who has traveled to an area in which malaria is endemic. Asymptomatic parasitemia can occur among persons who have been long-term residents of areas in which malaria is endemic.

Laboratory criteria for diagnosis

- Demonstration of malaria parasites in blood films

Case classification

Confirmed: an episode of microscopically confirmed malaria parasitemia in any person (symptomatic or asymptomatic) diagnosed in the United States, regardless of whether the person experienced previous episodes of malaria while outside the country

Comment

A subsequent attack experienced by the same person but caused by a different *Plasmodium* species is counted as an additional case. A subsequent attack experienced by the same person and caused by the same species in the United States may indicate a relapsing infection or treatment failure caused by drug resistance.

Permanent slides from all diagnosed and suspected cases should be sent to the State Laboratory.

Rocky Mountain Spotted Fever

reporting code = 08200
case report form: CDC 55.1 (1/01)
Tick-Borne Rickettsial Disease Case Report

Clinical description

A tickborne febrile illness most commonly characterized by acute onset and usually accompanied by myalgia, headache, and petechial rash (on the palms and soles in two thirds of the cases)

Laboratory criteria for diagnosis

- Fourfold or greater rise in antibody titer to *Rickettsia rickettsii* antigen by immunofluorescence antibody (IFA), complement fixation (CF), latex agglutination (LA), microagglutination (MA), or indirect hemagglutination antibody (IHA) test in acute and convalescent phase specimens ideally taken ≥ 3 weeks apart

OR

- Positive polymerase chain reaction (PCR) assay to *R. rickettsii*

OR

- Demonstration of positive immunofluorescence of skin lesion (biopsy) or organ tissue (autopsy)

OR

- Isolation of *R. rickettsii* from clinical specimen

Case classification

Confirmed: a clinically compatible case that is laboratory confirmed

Probable: a clinically compatible case with a single IFA serologic titer of ≥ 64 or a single CF titer of ≥ 16 or other supportive serology (fourfold rise in titer or a single titer ≥ 320 by Proteus OX-19 or OX-2, or a single titer ≥ 128 by an LA, IHA, or MA test)

Comment

Acute and convalescent sera should be acquired on all cases and sent to the State Laboratory. A copy of laboratory test results should accompany the case report form.

Yellow Fever

reporting code = 06090
case report form: N/A

Clinical description

A mosquito-borne viral illness characterized by acute onset and constitutional symptoms followed by a brief remission and a recurrence of fever, hepatitis, albuminuria, and symptoms and, in some instances, renal failure, shock, and generalized hemorrhages

Laboratory criteria for diagnosis

- Fourfold or greater rise in yellow fever antibody titer in a patient who has no history of recent yellow fever vaccination and cross-reactions to other flaviviruses have been excluded

OR

- Demonstration of yellow fever virus, antigen, or genome in tissue, blood, or other body fluid

Case classification

Confirmed: a clinically compatible case that is laboratory confirmed

Probable: a clinically compatible case with supportive serology (stable elevated antibody titer to yellow fever virus [e.g., ≥ 32 by complement fixation, ≥ 256 by immunofluorescence assay, ≥ 320 by hemagglutination inhibition, ≥ 160 by neutralization, or a positive serologic result by IgM-capture enzyme immunoassay]. Cross-reactive serologic reactions to other flaviviruses must be excluded, and the patient must not have a history of yellow fever vaccination.)

HUMAN CASE INVESTIGATION GUIDELINES

Priority

Human arboviral encephalitis case investigations should be initiated upon receipt.

Case Interview

- History of mosquito bites in 14 days prior to onset of symptoms
- Travel and activity history: travel outside county of residence, state, or country; occupation; hobbies (e.g., gardening, fresh water fishing, hunting); and other outdoor activities
- Environmental investigation: residence with screened windows, residence surrounded by vegetation or surface fresh water (lake, pond, etc)
- The recommended **case report form** can be found at:
http://www.doh.state.fl.us/Environment/hsee/arbo/reporting_links.htm

Disease Control Measures

A. Education

The risk of acquiring an arboviral illness is greatly reduced by taking precautions to limit exposure to mosquitoes. These should include the “5 D’s” for prevention:

Dusk and Dawn: avoid being outdoors when many mosquitoes are biting

Dress: wear clothing that covers skin

DEET (use mosquito repellents including DEET [N, N diethyl-*m*-toluamide] on skin and pyrethrins on clothing outdoors)

Drainage: check your home to rid it of standing water in which mosquitoes can lay their eggs

Additionally, elimination of breeding sites is key to prevention:

- Clean debris out of eaves, troughs and gutters.
- Remove old tires or drill drainage holes in those used in playgrounds
- Turn over or remove empty plastic pots.
- Pick up all beverage containers and cups.
- Check tarps on boats or other equipment that may collect water.
- Pump out bilges on boats.
- Replace water in birdbaths and pet or other animal feeding dishes at least once a week.
- Change water in plant trays, including hanging plants, at least once a week.
- Remove vegetation or obstructions in drainage ditches that prevent the flow of water.

B. Community Intervention

Medical alerts are issued by the DOH when surveillance systems indicate an increase in arboviral activity. CHDs should coordinate with Mosquito Control and Environmental Health Services to provide public information about “the 5 D’s: Dusk and Dawn, Dress, DEET, and Drainage”.

Laboratory Support

The Department of Health laboratories provide testing services for patients with clinical signs of arboviral encephalitis. These signs may include headache, fever, fatigue, dizziness, weakness and confusion. Due to the cross-reactivity between West Nile and other closely related flaviviruses, positive commercial

laboratory test results for antibodies to West Nile or other arboviruses should be confirmed by the DOH Bureau of Laboratories (i.e., positive specimens tested at private laboratories should be forwarded to the state laboratory for confirmation). Physicians should submit serum and cerebrospinal fluid samples to either the Tampa or Jacksonville Department of Health laboratories. In addition, if enterovirus is one of the differential etiologies, submission of an acute stool specimen or an acute throat swab is recommended. Even though a very early acute serum may be negative it is recommended that it be collected and submitted without waiting for the convalescent specimen. The convalescent specimen (drawn 2 weeks later) should be routinely sent to confirm negative and positive results.

A completed Florida Department of Health laboratory submission form should accompany all specimens <http://www.doh.state.fl.us/lab/labform.pdf> (please add the onset date of disease symptoms in the “Additional Tests/Comment” area). Proper collection, storage, labeling, and packaging of specimens are essential to ensure accurate test results, see directions at http://www9.myflorida.com/disease_ctrl/epi/httopics/arbo/3.htm. Serum and CSF specimens will be tested for antibodies to SLE, WN, EEE and dengue.

Algorithm for interpretation of laboratory results

Eastern Equine Encephalitis Serology

- 1a EEE IgM + **and**
- 2a IgG + (HI, IgG ELISA, IFA)..... Confirmed case
 - 2b IgG – Go to 3
 - 3a CSF IgM+..... Confirmed case
 - 3b Serum collected < 7 days post onset..... Probable case
Submit convalescent serum for confirmatory testing
 - 3c Serum collected > 7 days post onset..... Not a case
- 1b EEE IgM – **and**
- 4a IgG - Not a case
 - 4b IgG + Go to 5
 - 5a IgG+ in a single serum..... Go to 6
 - 6a IgG titer low..... Not a case
(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)
 - 6b IgG titer high..... Probable case
(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA)
 - 5b IgG+ in paired sera..... Go to 7
 - 7a Antibody titers in both sera are the same or < fourfold
difference Go to 8
 - 8a IgG titer low Not a case
(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)
 - 8b IgG titer high..... Probable case
(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA)
Submit convalescent serum for confirmatory testing
 - 7b Antibody titers in the two sera ≥ fourfold difference Confirmed case

Flavivirus Serology

Note: All specimens tested for flaviviruses at the DOH laboratories are tested for antibodies to multiple viruses (i.e. WNV, SLEV and DENV) before considered confirmed. Antibodies to all three viruses are often present in flavivirus positive sera. Specific antibodies to the virus causing the infection generally have the highest titers.

West Nile Virus Serology

1a	WNV IgM +	Go to 2
2a	IgG + by HI, IgG ELISA, or IFA assays.....	Probable case
	<i>PRNT assay needed to definitively distinguish WNV, SLEV and DENV ab</i>	
2b	IgG- by HI, IgG ELISA, or IFA assays.....	Go to 3
3a	CSF WN IgM +	Confirmed case
	3b Serum collected < 7 days post onset.....	Probable case
	<i>Submit convalescent serum for confirmatory testing</i>	
	3c Serum collected > 7 days post onset.....	Not a case
1b	WNV IgM –	Go to 4
4a	IgG -	Not a case
4b	IgG +	Go to 5
5a	IgG + single serum.....	Go to 6
6a	IgG titer low	Not a case
	(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)	
6b	IgG titer high	Probable Case
	(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA)	
	<i>Submit convalescent serum for confirmatory testing</i>	
5b	IgG + in paired sera	Go to 7
7a	Antibody titers in both sera are the same or < fourfold difference	Go to 8
8a	IgG titer low.....	Not a case
	(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)	
8b	IgG titer high.....	Probable case
	(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA)	
	<i>PRNT assay needed to distinguish SLEV, WNV and DENV ab</i>	
7b	Antibody titers in the two sera ≥ fourfold difference	Confirmed case

St Louis Encephalitis Virus Serology

1a	SLEV IgM +	Go to 2
2a	IgG + by HI, IgG ELISA, or IFA assays..... <i>PRNT assay needed to definitively distinguish WNV, SLEV and DENV ab</i>	Probable case
2b	IgG- by HI, IgG ELISA, or IFA assays.....	Go to 3
3a	CSF WN IgM +	Confirmed case
3b	Serum collected < 7 days post onset..... <i>Submit convalescent serum for confirmatory testing</i>	Probable case
3c	Serum collected > 7 days post onset.....	Not a case
1b	SLEV IgM -	Go to 4
4a	IgG -	Not a case
4b	IgG +	Go to 5
5a	IgG + single serum.....	Go to 6
6a	IgG titer low	Not a case
	(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)	
6b	IgG titer high	Probable Case
	(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA) <i>Submit convalescent serum for confirmatory testing</i>	
5b	IgG + in paired sera	Go to 7
7a	Antibody titers in both sera are the same or < fourfold difference	Go to 8
8a	IgG titer low.....	Not a case
	(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)	
8b	IgG titer high.....	Probable case
	(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA) <i>PRNT needed to distinguish SLEV, WNV and DENV ab</i>	
7b	Antibody titers in the two sera \geq fourfold difference	Confirmed case

Dengue Virus Serology

1a	DEN IgM +	Go to 2
2a	IgG+ by HI, IgG ELISA, IFA assays.....	Probable case
	• IgG titer low	Suspect primary infection
	(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)	
	• OR IgG titer high	Suspect secondary infection
	(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA)	
2b	IgG - by HI, IgG ELISA, IFA assays	Go to 3
3a	Serum collected < 7 days post onset.....	Probable case
	<i>Submit convalescent serum for confirmatory testing</i>	
3b	Serum collected > 7 days post onset.....	Not a case
1b	DEN IgM –	Go to 4
4a	IgG -	Not a case
4b	IgG +	Go to 5
5a	IgG+ in single serum.....	Go to 6
6a	IgG titer low.....	Not a case
	(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)	
6b	IgG titer high	Probable Case
	(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA)	
	<i>Submit convalescent serum for confirmatory testing</i>	
5b	IgG + in paired sera.....	Go to 7
7a	Antibody titers in both sera are the same or < fourfold difference between titers.....	Go to 8
8a	IgG titer low.....	Not a case
	(<1:320 by HI or <1:160 by PRNT or <1:256 by IFA)	
8b	IgG titer high	Probable case
	(>1:320 by HI or >1:160 by PRNT or >1:256 by IFA)	
	<i>PRNT assay required for definitively distinguish WNV, SLEV and DENV</i>	
7b	Antibody titers in the two sera ≥ fourfold difference	Confirmed case

Tickborne Illness

Tickborne illnesses of concern in Florida include Ehrlichiosis, Lyme disease, and Rocky Mountain spotted fever. Lyme disease is mostly localized to states in the northeastern, mid-Atlantic, and upper north-central regions (Connecticut, Rhode Island, New York, Pennsylvania, Delaware, New Jersey, Maryland, Massachusetts, and Wisconsin). RMSF cases have been largely reported from the south-Atlantic region and the western south-central region; few cases are reported from the Rocky Mountain region.

Priority

Human tickborne illness case investigations should be started within three days of receipt.

Case Interview

- History of tick bite in 14-21 days prior to onset of symptoms
- Travel and activity history: occupation, hobbies (e.g., camping, hunting, other outdoor activities, especially in woodsy areas)
- Environmental investigation: residence surrounded by woods or forest (ticks especially like a grass/forest border from which to quest or wait for the next animal or human to bush by or approach), deer or rodents on property
- Copies of the case report forms for Ehrlichiosis, Lyme Disease and RMSF can be found on Bureau of Epidemiology's website http://www.doh.state.fl.us/disease_ctrl/epi/topics/crforms.htm.

Disease Control Measures

A. Education

The risk of acquiring a tickborne illness is greatly reduced by taking precautions to limit exposure to ticks.

- Avoid tick habitats if possible
- If exposure to tick habitats cannot be avoided, when outdoors in a tick area, cover up by wearing shoes, socks, long pants and long-sleeved shirts (light colored clothing preferred for spotting ticks)
- Use insect repellent containing DEET according to the manufacturer's directions
- Perform daily tick checks

B. Community Intervention

- Control tick populations in yards and on pets
- Protect pets from ticks by consulting with your veterinarian

Laboratory Support

Laboratory criteria differ by tickborne illness and may be based upon paired sera antibodies, or IgM and IgG antibody detection. Please refer to the Bureau of Epidemiology Surveillance Case Definitions for Select Reportable Diseases in Florida for additional information on laboratory criteria for reporting Ehrlichiosis, Lyme, and RMSF http://www9.myflorida.com/disease_ctrl/epi/surv/CaseDefJune2003.pdf. Also refer to "Notice to Readers Recommendations for Test Performance and Interpretation from the Second National Conference on Serologic Diagnosis of Lyme Disease" published in the MMWR, August 11, 1995/44(31) 590-591 <http://www.cdc.gov/mmwr/preview/mmwrhtml/00038469.htm>. The Centers for Disease Control and Prevention (CDC) recommends laboratories follow a 2 step process for detection of lyme antibodies, an ELISA or IFA screen with IgM or IgG confirmation by Western Blot (IgM if serum is taken < 4 weeks since symptoms began, IgG if serum is taken > 4 weeks since symptoms began).

WEST NILE VIRUS MOSQUITO-POOL PROTOCOL

1. Mosquito collection techniques
 - 1.1. Traps:
 - 1.1.1. CDC (with or without CO²),
 - 1.1.2. Gravid, ABC light traps (with CO²), MM-X traps (a.k.a. pickle jar) (with CO²),
 - 1.1.3. Lardcan, or
 - 1.1.4. Mosquito Magnet traps
 - 1.2. Traps may be set anywhere West Nile (WN) virus transmission is suspected to be ongoing. Remember that arboviral transmission can be extremely focal in widely dispersed habitats. So other trap sites and collection techniques should also be considered including ground aspirator collections at mosquito daytime resting sites, avian roosts and areas of past virus activity.
 - 1.3. Maintain accurate and detailed nightly records for each collecting bags and each resulting mosquito pool.
 - 1.4. Priority: ornithophilic and opportunistic mosquitoes;
 - 1.4.1. *Culex*
 - 1.4.2. *Culiseta*
 - 1.4.3. *Mansonia*
 - 1.4.4. *Coquillettidia*
 - 1.4.5. *Aedes*
 - 1.4.6. *Ochlerotatus*
2. Mosquitoes should be live or recently (< 2 hr) dead, non-fed or gravid females only.
Do not pool blood-fed mosquitoes because, if positive, it is impossible to tell whether the virus originated in the mosquito or in the blood meal.
3. Sample Processing:
 - 3.1. Hold samples on wet ice in field or transport traps in coolers to laboratory
 - 3.1.1. Do not use dry ice to kill or anesthetize collections because the carbon dioxide acidifies the sample and may kill the virus, thus interfering with tests designed to isolate live virus. However, it *is* desirable to ship mosquitoes that are sealed within proper tubes to the Tampa Laboratory on dry ice (see instructions in section 3.5).
 - 3.1.2. Make sure mosquitoes are kept alive by keeping them in a humid environment with access to cotton balls soaked with 5% sugar water.
 - 3.1.3. Once mosquitoes are killed they must be kept in a freezer maintained at -70° C or colder.
 - 3.3. Use a chill table to sort the specimens. Triethylamine (TEA) can also be used to anesthetize the insects for the sorting process.
 - 3.4. Group female mosquitoes into pools of 50 individual mosquitoes by species, site and week (or night) of collection. Be careful not to contaminate the sample by including loose body parts (e.g. legs) belonging to other mosquito pools.

- 3.5. **Do not combine mosquitoes or mosquito species trapped on different nights, different sites, or in different types of traps at the same site.**
- 3.6. Make sure each mosquito pool is clearly and accurately labeled with a unique identifier number. This information plus any notes or comments for each pool should appear on a master data sheet, which is copied and maintained in two separate locations. Information on the pool should include:
 - 3.5.1. mosquito species
 - 3.5.2. number of specimens
 - 3.5.3. mosquito data (sex and empty or gravid for females)
 - 3.5.4. collection date
 - 3.5.5. collection location
 - 3.5.6. collection method (attractant trap type or non-attractant collection; if traps used, note attractant used as this indicates bias for particular age classes)
- 3.6. Accurate species identification is essential. If you are unsure of the species identification do not guess. Either have the specimen accurately identified or discard it. Unidentified pools will be not be tested by the Tampa Laboratory.
- 3.7. Label tubes (preferably 2.0 ml plastic, snap-cap microcentrifuge tubes (Fisher Cat # 02-681-258) with the unique identification number or with the following information: species name and number, site, collection date, numbers of mosquitoes. Seal the tube with plastic film (or plastic electrical tape) and store it at -70° C. A proper seal is essential to prevent intrusion by carbon dioxide gas if the specimens are shipped on dry ice!_Maintain accurate records.
- 3.8. Complete the “Arbovirus Surveillance, virus isolation” form and send with the submitted pools to DOH Tampa Laboratory
 - 3.8.1. Drive to DOH Tampa Laboratory or overnight mail on dry ice.
 - 3.8.2. Laboratory address:
Dr Lillian Stark
Bureau of Laboratories
Tampa Branch Laboratory
3602 Spectrum Blvd
Tampa, FL 33612
Tel: 813-974-5990

4. Sample assay/reporting

- 4.1. Samples are screened in a molecular assay (TaqMan RT-PCR) for WN virus.
 - 4.1.1. Pools positive for WN virus are reported by email to the submitter.
 - 4.1.2. When molecular screening is completed, a report is mailed to the submitter.
- 4.2. Samples are inoculated onto cell cultures for arbovirus isolation.
 - 4.2.1. When an isolate is detected it is identified by RT-PCR using multiple primer sets. and probes. Gene sequencing may be performed.
 - 4.2.2. Virus isolates are reported by email to the submitter.
 - 4.2.3. When isolation attempts are completed, a report is mailed to the submitter.
5. To benefit arboviral surveillance programs, mosquitoes should be pooled and shipped to the Tampa Laboratory within 24 hours of collection. In addition, the shipments need to arrive at the Laboratory

on a weekday to make sure staff is available to process the specimens. Results will be reported back to the collector within two weeks.

MALARIA

Introduction

Malaria is one of the world's greatest public health problems. Approximately 300 million of the world's population are infected each year and between 1 and 1.5 million people die from malaria annually. Although malaria is no longer endemic in Florida, it is often seen in travelers and unusual locally acquired cases can be seen in the state.

Human malaria is caused by four species of protozoan parasites of the genus *Plasmodium*: *P. vivax*, *P. falciparum*, *P. malariae*, and *P. ovale*. All four are transmitted from person to person via the bite and blood-feeding behavior of mosquitoes of only the genus *Anopheles*. Thus, part of the complex life cycle occurs in humans and part in the mosquito.

Vector

In Florida, there are 13 *Anopheles* species, all of which are potentially capable of transmitting malaria:

Anopheles quadrimaculatus A, B, C and D

- Principal malaria carrier.
- Found in every county, more abundant in northern Florida.
- Breeds in alkaline ponds, lakes and gum swamps in the limestone and red clay regions of northern and western Florida.

An. crucians

- Breeds in acid ponds and cypress swamps.

An. punctipennis

- Breeds in winter in slow-flowing alkaline streams of northern and western Florida.

0 *An. perplexens*

- Rare mosquito found in north central Florida.

An. atropos and *An. bradleyi*

- Breeds in salt marshes.

1 *An. albimanus*

- Very rare species.
- Breeds in sunlit pools on the Florida Keys.
- Major malaria vector in Central America.

2 *An. walkeri*

- More common in central Florida.
- Breeds in heavily vegetated lakes.

An. georgianus

- Rare species.
- Breeds in seepage areas.

3 *An. barberi*

- Breeds in tree holes.

Epidemiology

Although now rare in the United States, malaria was once the major scourge of Florida (both *P. vivax* and *P. falciparum*), occurring in all 67 counties. Data collected since 1917 from the Bureau of Vital Statistics (Provost 1946, unpublished) showed 24 counties with annual death rates from malaria of 100 per 100,000; eight had rates above 200; and Dixie County, in 1930, above 300. According to the usually accepted ratio of 200 malaria cases per death, these rates meant 20, 40, and 60% of the populations involved had malaria. The 24 counties having the highest rate of malaria in Florida and the U.S. were Dixie, Taylor, Jefferson, Lafayette, Wakulla, Gilchrist, Madison, Citrus, Levy, Hernando, Gadsden, Suwannee, Leon, Jackson, Calhoun, Franklin, Okeechobee, Hamilton, Washington, Pasco, Sumter, Columbia, Holmes and Liberty. Malaria morbidity reports for Florida show a steady decrease since 1934 with no large outbreak since 1937. This reduction in malaria incidence was probably due to adult mosquito sprays, improved housing included screening, use of repellents, agricultural and other drainage practices and the use of anti-malarial drugs.

Until recently, the last case of malaria from the bite of a naturally infected mosquito occurred in 1948. In June 1990, Florida had its first case of human malaria (*P. vivax*) in 42 years, acquired presumably through the bite of a mosquito in Gulf County that became infected after biting a migrant worker with malaria. Two induced cases of *P. falciparum* occurred in Broward County in 1996 and were probably related to iatrogenic spread in a hospital setting where a patient was being treated for imported malaria infection. Two cryptic cases occurred in Palm Beach County also in 1996 and resulted in *P. vivax* infection. One of these cryptic cases was in a homeless male and the other was in a resident living in a nearby area. The largest *P. vivax* outbreak in recent Florida history (with seven cases) occurred in Palm Beach County in 2003.

In the Americas, over 1 million cases occur annually and approximately 30% of its population reside in areas suitable for malaria transmission. The largest number of cases are reported from Brazil, which accounted for 50% of the total in 1994, followed by the Andean countries, which reported 29% of all cases. The CDC received reports of 1,800 cases in 1996 for the U.S. The number of cases in the U.S. has been gradually increasing from the early 1970s and may represent increasing cases from migrants and increased travel among U.S. citizens. Of the less than 100 Florida cases per year reported in recent years, more cases originated from exposure in Central American countries than any other area.

Clinical Course

In humans, the symptoms will vary depending on the malaria species, but the initial attack may start with lassitude, headache, anorexia, occasional nausea and vomiting. The fever is comprised of a cold stage (shivering and a feeling of intense cold), a hot stage (distressing heat, dryness, burning, intense headache, nausea, and vomiting) and finally a profuse sweating stage. The typical attack often begins in the early afternoon and lasts from eight to twelve hours. Persons experiencing these symptoms and having been in an area with malaria are encouraged to see a doctor immediately.

P. vivax occurs throughout most of the temperate zone, large areas of the tropics, and less commonly in tropical Africa. Severity of the primary attack ranges from mild to severe, usually not resulting in death. *P. falciparum* is generally confined to tropical or subtropical regions and is particularly severe and often fatal in infants, young children and in non-immune persons. *P. malariae* is frequently named "quartan malaria" because the fever recurs on the fourth day after a two-day interval. The fevers of the other three malaria species recur on the third day after a one-day interval. *P. malariae* occurs over both tropical and sub-tropical areas. The disease is less severe, but may have a long persistence. *P. ovale* is similar to *P. vivax* malaria, but with a prolonged latency and generally milder clinical symptoms. It is most common in West Africa.

Specific characteristics

Vivax malaria

Clinical:

- Incubation period 12-17 days (9-10 months recorded)
- Primary attack (8-10 hours duration)
- Sudden, shaking chill often for several hours, headache, back pain, nausea, malaise
- Irregular fever during the first 2-4 days up to 104-105 degrees
- Fever terminates by crisis with drenching sweat, up to several hours
- Series of fevers every 48 hours with diminishing intensity for 2 weeks
- Two-week latent period
- Secondary attacks (less intense) for 2 months
- Six- to nine-month latent period
- Long-term relapses - 2.5-3 years

Pathology:

- Infects new red blood cells, red cell destruction leads to anemia
- Enlarged spleen, pulp tarry, malphigian bodies pale gray, malaria pigment within reticulo-endothelial cells
- Congested and enlarged liver, destruction of the bile canaliculi
- Granular casts and fatty degeneration in kidneys
- Infected RBCs are sticky and adhere to capillary, hemorrhages, tissue anoxia and electrolyte imbalance

Falciparum malaria

Clinical:

- Incubation period 9-14 days
- Headache, back pain, prostration, chill
- Fever irregular, and no distinct periodicity, sweating may be present even when fever is low, higher temperature up to 105-110 degrees F
- Pulse and respiration rates are rapid
- Nausea, vomiting and diarrhea increase, frequently a cough
- Cerebral manifestations of excitation, depression, behavioral changes with psychotic tendencies, coma without hyperpyrexia
- Bilious form - nausea, vomiting, gastric distress, jaundice
- Algid form - high internal heat, body cold and clammy
- Choleraic form - stools loose ("rice water")
- Severe dehydration and anemia
- If untreated, "pernicious malaria" may develop suddenly
- Frequent recrudescence during first month
- Radical cure in about 10 months

Pathology:

- Infects all red blood cells
- Few parasites may be present
- Spleen and liver enlargement
- Acute hemolysis of erythrocytes (hemoglobinuria) with dark, mahogany-red urine (blackwater fever)
- Renal failure

Malariae malaria

- Clinical symptoms similar to vivax, but may be more severe
- Untreated infections may have relapses 30-50 years later

Ovale malaria

- Clinical symptoms similar to vivax
- Spontaneous recovery common, fewer relapses

Surveillance Issues

Imported malaria will continue to be an issue from travelers and visitors to Florida, including migrant workers. Locally acquired cases are possible due to the presence of *Anopheles* throughout the state in the presence of parasitic human hosts. Surveillance and investigation of reported cases will continue to be important. The surveillance data will be optimized by the following activities.

- Remind physicians and public health workers regularly about the importation of malaria among travelers and visitors, including migrant workers, and the danger of not clinically diagnosing malaria from more common febrile illnesses and immediately reporting all confirmed cases.
- Obtain slides and conduct thorough investigations of all cases with special attention to finding secondary cases and preventing further disease.
- Inform all public health officials including state and county health officers, mosquito control directors, and the Director of the FMEL of all imported malaria cases by county in Florida.
- Survey and map annually all actual and potential anopheline larval breeding sites in the district. Annually map anopheline adult distribution and record the seasonal abundance collections in the county. Be informed of all imported and introduced malaria in the county and Florida.

Any case that is not readily explained by foreign travel or visitors (including migrant workers) is strongly suggestive of local transmission. When a case of malaria has been identified, the public is warned to report any fever of unknown origin to their physician or county health department. A blood film and purple-top tube are submitted for hemoparasitologic analysis of all fever cases suspected of having malaria. It is important that the specimens are collected before treatment is initiated. Depending on the number of cases (at least two), the county health department may conduct a survey of migrant workers and local residents (family and neighbors) in the immediate area where the malaria cases occurred. The case definition for malaria can be found in Appendix D.

Depending on circumstances such as abundance of vectors, human population density in the area, number of suspected human cases, etc., mosquito abatement measures may be initiated. Abatement responses are coordinated with DACS Bureau of Entomology and Pest Control.

DENGUE AND YELLOW FEVER

Introduction

Dengue (DEN) and yellow fever (YF) are two important mosquito-borne diseases that have historically plagued Florida, although not for more than 50 years. Yellow fever is the result of a single virus species that typically causes profound hemorrhagic disease, which is often fatal. The syndromes collectively referred to as "dengue" and dengue hemorrhagic fever (DHF) are caused by any of four closely related virus subtypes. Classical dengue (so-called "break-bone fever") is a painful, debilitating febrile disease that is rarely fatal. This illness is characterized by abnormal vascular permeability, hypovolemia and abnormal blood clotting mechanisms. Dengue hemorrhagic fever-dengue shock syndrome (DHF-DSS) is a group of severe hemorrhagic symptoms that occur principally in children but may also occur in adults. In those with severe disease, shock is the predominant sign. Case fatality rate can be as high as 40-50% untreated, but can be drastically lowered with appropriate fluid therapy. Encephalitis is a rare consequence of dengue infection. The pathogenesis and risk factors associated with DHF-DSS are controversial but appear to be related to second or greater infection with dengue serotypes.

In past Florida epidemics, the sole vector of both DEN and YF was undoubtedly *Aedes aegypti*. The recent arrival of *Aedes albopictus* to many parts of Florida is disturbing, since this species is an important vector of DEN viruses in Asia. *Ae. aegypti* is highly domesticated, and almost exclusively utilizes artificial containers as larval habitats. In contrast, *Ae. albopictus* is fundamentally a treehole- and leaf axil-dwelling species that is secondarily an artificial container dweller.

In parts of Asia having both DEN vectors, there is a tendency for urban DEN cases to be *Ae. aegypti*-transmitted, while suburban and rural cases are *Ae. albopictus*-transmitted. Major DEN epidemics have also occurred in large Asian cities inhabited by *Ae. albopictus*, but not *Ae. aegypti*. Most experimental comparisons have shown *Ae. albopictus* to be a more efficient vector of DEN viruses than *Ae. aegypti*. North American strains of *Ae. albopictus* have been shown competent to serve as vectors of YF virus as well, and the biology of this species offers the potential to establish a "sylvatic" transmission cycle in Florida. Possibly because the geographic ranges of YF virus and *Ae. albopictus* have only recently begun to overlap, there is no documented evidence of YF transmission in the Americas by this species.

DEN and YF have become increasingly common diseases in the Caribbean, Central America, the Pacific and South America during the past two decades. Humans are the only important vertebrate hosts of DEN viruses. So-called "urban" YF involves transmission between humans and *Ae. aegypti*, and is manifest in large epidemics. Puerto Rico and other Caribbean islands experience DEN epidemics annually. Florida's proximity to the Caribbean suggests that outbreaks of DEN are likely to recur in the state, despite their absence since the 1930's. A focus of YF transmission is probably less likely to appear in Florida. It would be possible for either DEN or YF viruses to be imported into Florida by inadvertent transport of infected mosquitoes. However, the occurrence of at least one vector species in many parts of Florida increases the probability that Florida *Ae. aegypti* or *Ae. albopictus* females will be exposed to imported DEN or YF viruses after feeding on viremic travelers returning from the Caribbean or Central America. For DEN and YF case definitions see Appendix D.

Surveillance

Importation and establishment of DEN or YF viruses in Florida will occur unpredictably, perhaps not for many years. Unfortunately, isolated outbreaks of classic DEN typically grow to involve hundreds of cases before local health authorities correctly identify them. Minimal surveillance in Florida involves annual notification of physicians and public health authorities of the possibility of DEN (or YF) cases in Florida. Clinical differentiation of these exotic diseases from more common febrile illnesses may be difficult. Therefore, immediate submission of sera from all suspect cases to the DOH viral serology laboratory is needed for confirmation. Humans will clearly play the role of "sentinel host" for imported and/or locally transmitted DEN, DHF-DSS, or YF in Florida.

Appropriate, recurring education of medical and public health personnel is a theoretically effective means of minimizing the impact of an introduction of one of these viruses. Although an effective vaccine has long been available for YF virus, widespread immunization of the resident population to preclude establishment of imported YF would not be appropriate or feasible. There are no reliable vaccines available for any of the dengue viruses.

Recognition of a focus of DEN or YF transmission in Florida requires an immediate and energetic response by local mosquito control personnel to reduce exposure of residents to *Ae. aegypti* and *Ae. albopictus* vectors. This involves treatment or removal of all container habitats found in the area. Ground level adulticiding may be appropriate, but aerial adulticiding is generally thought to be ineffective in the control of dengue outbreaks. Vigorous public education through the news media encourages residents to take appropriate personal protection measures and assist in the effort to eliminate artificial container habitats.

Identification of a focus of local DEN or YF transmission anywhere in Florida elicits immediate notification of physicians and public health workers due to the potentially explosive nature of these diseases.

Since neither disease is currently endemic, ANY case of DEN or YF that is not readily explained by recent foreign travel is strongly suggestive of local transmission. In such a situation the threat of additional cases in the near-term is substantial. Likewise, there is the possibility that virus may become endemic if local populations of *Ae. aegypti* or *Ae. albopictus* are large. As a practical matter, a single human case that is not imported but of local origin, elicits an immediate "medical alert" (see Chapter 4 for definition of and response to a medical alert).

TICK REMOVAL/STORAGE AND IDENTIFICATION AFTER TICK-BORNE DISEASE DIAGNOSIS

Ticks suspected as potential disease vectors in the southeastern US are among the following:

Ixodes scapularis

Common name: Black-legged Tick



Adult Female

Seasonal Abundance: Larvae and Nymphs: April - August
 Adults: September - May

Hosts: Larvae and Nymphs: Reptiles (skinks and snakes), birds and to much lesser degree rodents.
 Adults: Larger animals including cattle and humans.

Amblyomma americanum

Common Name: Lone Star Tick



Adult Female

Seasonal Abundance: Larvae: June - November
 Nymphs: February - October
 Adults: April - August with peaks in July

Hosts: Larvae and Nymphs: Small mammals and birds. Do not feed on rodents.
 Adults: Deer, cattle and humans.

Amblyomma maculatum

Common Name: Gulf Coast Tick



Adult Female

Seasonal Abundance: Larvae: June - September
Nymphs: February - October
Adults: June - September

Hosts: Larvae and Nymphs: Ground birds and small rodents
Adults: Larger mammals including cattle, deer, dogs and humans

Dermacentor variabilis

Common Name: American Dog Tick



Adult Female

Seasonal Abundance: Larvae: July - February
Nymphs: January - March
Adults: March - September

Hosts: Larvae and Nymphs: Almost exclusively small rodents, particularly mice and cotton rats
Adults: Large variety of mammals and humans

Tick Removal and Storage for Possible Identification

Ticks are best removed using spoon-type devices to wedge the tick off without touching it (for example, Ticked Off). Without such a device, ticks can be removed by firmly grasping the tick at the point of attachment with tissue-covered fingers and applying slow, steady traction.

Tick identification may be of interest to the person or the physician; however, it will not predict whether or not the person will become infected with a particular disease. For this reason, many entomologists suggest using tick identification **as a supplement to diagnosis by a physician of a tick-borne disease.**

Ticks may be placed separately in small jars or vials stuffed to about 1/2" depth with paper towels and sealed with a top containing air holes. The containers should be placed inside a zip-lock plastic bag containing moistened cotton balls or paper towels and stored in a refrigerator. Ticks will survive for several weeks to months using this technique. The bag should be labeled with: name of patient, date, location and contact person's phone number. Specimens must be mailed along with a completed tick submittal form (below).

Tick Identification Submittal Form

After the physician has made a presumptive diagnosis of a tick-borne disease, a completed copy of this form should be sent along with the tick specimen to:

Dr. John P. Smith
John A. Mulrennan, Sr. Public Health Entomology Research and Education Center
Florida A & M University
4000 Frankford Avenue
Panama City, Florida 32405

Date: _____ Submitter's Name: _____ Phone: _____

Submitter's address: _____

Reason for submitting tick: _____

Patient's Name/Address/Phone Number: _____

Patient's Age: _____ Sex: _____

Where tick was acquired: City _____ State _____

Specific location/address _____

MODEL MEDIA KIT**St. Louis Encephalitis Public Information Efforts**

During the summer of 1997, activity among sentinel chicken flocks indicated the potential for widespread human cases of SLE. Because personal prevention of mosquito bites is known to reduce the risk of arboviral infection, the Department of Health (DOH), county health departments and Mosquito Control Agencies undertook many activities to more adequately inform the public about the prevention of this dangerous disease. Three main public health messages were widely disseminated. The public was warned to: (1) minimize outdoor activities from dusk to dawn; (2) but, when outdoors during these hours, cover up with clothing; and (3) use mosquito repellents, as directed, on exposed skin. To draw attention to the potential danger and reinforce suggested preventive measures, the DOH issued a medical alert for 27 central and southern Florida counties. Significant media attention was generated by this alert and was used by the department both to reiterate the preventive messages and to communicate current viral activity in humans and chickens. During the season, nine cases of human illness, including one death, were recorded.

In an attempt to assess the effectiveness of the DOH's media campaign, several questions were appended to the Behavioral Risk Factor Surveillance System surveys for November and December [the alert was in place from August through mid-December]. Results of the survey follow: A total of 468 persons completed the SLE section of the survey, of which 184 were male and 284 were female. The mean age of respondents was 51 years. There were 286 respondents who lived in a county that had been placed on SLE alert. There were no differences between alert and non-alert counties with respect to age, sex or race/ethnicity.

Respondents were asked if they currently took any precautions to prevent mosquito bites. Of those answering the survey, 67% in alert counties and 51% in non-alert counties reported currently taking precautions ($p=0.001$). In alert counties, 93% of respondents reported having heard (or read) SLE messages, compared to 75% in non-alert counties ($p=0.001$). Of those who received SLE messages, 72.5% used some kind of anti-mosquito precaution compared to 45.3% of those who did not receive SLE messages ($p=0.001$). Television and newspapers were the most common sources of information on SLE. There were 86% of respondents in alert counties and 74% in non-alert counties who reported receiving SLE information from television ($p=0.002$); and 55% of respondents in alert counties and 39% in non-alert counties who reported receiving information from the newspaper ($p=0.003$). Of respondents who reported receiving SLE information, 41% reported taking additional precautions against mosquito bites after hearing the messages. In alert counties this number was 49%, and in non-alert counties, 27% took additional precautions ($p=0.001$). The most common preventive measures included the following: limiting outdoor activities (45.8% in alert counties versus 17.6% in non-alert counties, $p=0.001$); wearing insect repellent (44.8% in alert counties vs. 38.5% in non-alert counties, $p=0.2$); and wearing long pants and long sleeves (26.9% in alert counties vs. 10% in non-alert counties, $p=0.001$).

Widespread dissemination of these important preventive messages did not require large expenses for media airtime or print space by public agencies, but seemed to have been widely heard and practiced. Press releases, websites, toll-free hotlines and interviews with media representatives were commonly used to increase awareness of the message. These efforts probably prevented a large amount of morbidity as well as mortality during the 1997 SLE season and could be applied to other vector-borne diseases.

SAMPLE PRESS RELEASE for MEDICAL ALERTS

DRAFT

August XX, 2003

CONTACT: name

XXX-XXX-XXXX

MEDICAL ALERT ISSUED FOR XXXXXXXX COUNTY

--Human Case of (West Nile Virus, EEE, SLE, Dengue) Confirmed--

Today, County Health Department Director/Administrator (Dr.) **XXXXXX XXXXXXX** announced that Florida Department of Health (DOH) Secretary John O. Agwunobi, M.D., M.B.A., has issued a medical alert for **XXXXXX** County after a human case of (West Nile (WN) virus encephalitis/fever, EEE, Dengue, Malaria) was confirmed in a XX-year-old (fe)male resident.

<Insert symptoms here> Physicians should contact the County Health Department if they suspect an individual may meet the case definition for a mosquito-borne illness. DOH laboratories provide testing services for physicians treating patients with clinical signs of mosquito-borne disease.

DOH continues to advise the public to remain diligent in their personal mosquito protection efforts. These should include the "5 D's" for prevention:

- **Dusk and Dawn** -- *Avoid being outdoors when mosquitoes are seeking blood, for many species this is during the dusk and dawn hours.*
- **Dress** -- *Wear clothing that covers skin.*
- **DEET** -- *When the potential exists for exposure to mosquitoes, repellents containing DEET (N,N-diethyl-meta-toluamide, or N,N-diethyl-3-methylbenzamide) are recommended. Products with concentrations up to 30% DEET are generally recommended for most situations. (It is not recommended to use DEET on children less than 2 months old. Instead, infants should be kept indoors or mosquito netting used over carriers when mosquitoes are present). If additional protection is necessary, apply a permethrin repellent directly to your clothing. Always read the manufacturer's directions carefully before you put on a repellent.*
- **Drainage** -- *Check your home to rid it of standing water in which mosquitoes can lay their eggs.*

Elimination of breeding sites is one of the keys to prevention.

Tips on Eliminating Mosquito Breeding Sites

Clean out eaves, troughs and gutters.

Remove old tires or drill holes in those used in playgrounds to drain.

Turn over or remove empty plastic pots.

Pick up all beverage containers and cups.

Check tarps on boats or other equipment that may collect water.

Pump out bilges on boats.

Replace water in birdbaths and pet or other animal feeding dishes at least once a week.

Change water in plant trays, including hanging plants, at least once a week.

Remove vegetation or obstructions in drainage ditches that prevent the flow of water.

DOH continues to conduct statewide surveillance for mosquito borne illnesses, including West Nile (WN) virus, Eastern Equine Encephalomyelitis (EEE), St. Louis Encephalitis (SLE), malaria, and dengue. Residents of Florida are encouraged to report dead birds via the Web site <http://www.wildflorida.org/bird>. For more information on mosquito-borne illnesses, visit DOH's Environmental Health website at <http://www.doh.state.fl.us/Environment/hsee/arbo/index.htm>, call the West Nile Virus Hotline at 1-888-880-5782, or call your local county health department.

**Florida Department of Health
Public Service Announcement**

How to Protect Yourself From Mosquito-transmitted Encephalitis

St. Louis encephalitis (SLE)/ West Nile (WN)/ Eastern Equine Encephalomyelitis (EEE) is a serious disease people can get from mosquito bites. Follow the 5 D's for arbovirus prevention::

- Dusk and Dawn (Avoid being outdoors when mosquitoes are most active.)
- Dress (Cover your skin with clothing.)
- DEET (Use mosquito repellent on bare skin and clothing.) and
- Drain (Remove standing water in which mosquitoes can lay their eggs.)

Play it safe and keep mosquitoes from putting the bite on you!

SLE MEDICAL ALERT

The onset of St. Louis encephalitis (SLE) usually occurs within 4-21 days (onset of WN virus encephalitis (WN) usually occurs within 2-15 days...) after being bitten by a mosquito carrying the virus. Symptoms include fever, headache, stiff neck, dizziness, weakness, confusion, swelling of the brain, and, in the most severe cases among the elderly, coma and death. See your physician if you feel you have this disease.

Residents and visitors, especially the elderly, in alert counties are advised take basic precautions to reduce their exposure to mosquitoes and prevent encephalitis infection.

Remember the 5 D's for arbovirus prevention:

- Dusk and Dawn (Avoid being outdoors when mosquitoes are most active.),
- Dress (Cover your skin with clothing.),
- DEET (Use mosquito repellent on bare skin and clothing.) and
- Drain (Remove standing water in which mosquitoes can lay their eggs.)

Note: While the Department of Health has long-recommended that residents and visitors to counties under medical alerts limit their outdoor activities during dusk through dawn, the department is not recommending a large-scale ban of evening activities. Residents are advised to follow the common-sense precautions, such as wearing mosquito repellent and long-sleeve shirts and long pants, to make their time outside safer.

St. Louis Encephalitis (SLE)/ West Nile (WN) Questions and Answers

What is (St Louis Encephalitis, West Nile)?

St. Louis Encephalitis (SLE)/ West Nile (WN) is a mosquito-borne viral disease that causes inflammation (swelling) of the brain. In an average year, Florida has from one to 10 cases of SLE. Several large outbreaks involving as many as 200 cases-have occurred in Florida in recent decades./ More than 125 cases have been reported since West Nile virus was first detected in the state in 2001.

What are the symptoms of SLE/WN?

Many infections with SLE/ WN are inapparent but when symptoms occur they can range from fever with headache to coma. Other symptoms include: fatigue, dizziness, weakness and confusion.

Who is at risk of contracting SLE/ WN?

SLE virus/ WN virus is maintained in a bird-mosquito cycle. People may get the virus by being bitten by infected mosquitoes. While the virus can affect anyone, it has its greatest impact on people over the age of 50.

Is there a vaccine for SLE/ WN?

No. There is no vaccine because the virus occurs in humans so infrequently.

How can a person prevent infection?

Prevention is the key. The best way to avoid infection is to avoid getting mosquito bites.

Recommendations are:

- Check residential screening, including porches and patios
- Avoid outdoor activities between dusk and dawn
- If you must be outdoors when mosquitoes are active, cover up by wearing shoes, socks, long pants and shirts and use mosquito repellent on skin that will be exposed.
- Eliminate stagnant water in any receptacles in which mosquitoes might breed

When was the last outbreak of SLE in Florida?

In the fall of 1997, 9 contracted SLE. Florida's largest epidemic of SLE occurred in 1990, with 223 cases and 10 fatalities in central and southern areas of the state./ WN N/A

How do we know that SLE/ WN is in an area and that people might become infected?

Mosquito Control Districts located throughout the state continually monitor the distribution and density of mosquito populations known to carry the SLE/ WN virus. In many areas, these agencies and county health departments also keep chicken flocks and monitor these chickens for evidence of exposure to SLE/ WN virus.

How is this information communicated to the public?

State and county agencies monitor this information regularly and issue warnings to the public when mosquito populations are large and virus activity is detected.

What parts of the State of Florida are most at risk?

Historically, SLE virus has been detected throughout the state although outbreaks have tended to occur more in Central Florida from coast to coast./ the WN virus outbreak is occurring statewide.

What measures are government agencies taking to protect the population?

Mosquito control activities targeted against adult and larval populations have increased as a direct response to the reports of increased SLE/ WN activity. In addition, a number of press releases and public education activities have been undertaken to increase awareness of personal protective measures.

Malaria Questions and Answers

What is malaria?

Malaria is a serious disease caused by a parasite and carried by mosquitoes.

How do you get malaria?

You get malaria from the BITE of an infected MOSQUITO.

People who travel to a foreign country where malaria is common have the highest risk for malaria. However, it is possible to get malaria in Florida. The best way to protect yourself from malaria is to not get bitten by mosquitoes.

What are the signs and symptoms of malaria?

Symptoms of malaria include fever and flu-like illness, including chills, headache, muscle aches, and tiredness. Loss of appetite, nausea, vomiting, and diarrhea may also occur. Malaria may cause anemia and jaundice (yellow coloring of the skin and eyes) because the malaria parasites destroy red blood cells.

How soon will a person feel sick after being bitten by an infected mosquito?

For most people, symptoms begin 10 days to 4 weeks after infection.

What is the treatment for malaria?

Malaria **CAN** be treated and cured by the right prescription medications. A doctor **MUST** guide treatment.

How can you lower your chances of getting malaria?

The good news is that you **CAN** lower your chances of getting malaria and other diseases spread by mosquitoes by following the five “D’s” and one “S”:

- Between **Dusk** and **Dawn** (including nighttime): Avoid or limit outdoor activities as much as possible during the dusk, dawn and nighttime hours to avoid being bitten by mosquitoes. Nighttime is when mosquitoes that spread malaria bite.
- **Dress**: Wear long pants, long-sleeved shirts and socks outdoors during dusk, dawn and nighttime hours.
- **DEET**: Use an insect repellent containing DEET on exposed skin and follow the directions on the label.
- **Screens**: Close windows at night, or install screens in windows and doors if left open at night. If you do not live in a screened or air-conditioned house, sleep under a mosquito bednet that has been dipped in an insecticide containing permethrin.

If you think you have malaria?

See a doctor. Malaria can be treated.

Note: Products with concentrations up to 30% DEET are generally recommended for most situations.

DEET Information

Source: Centers for Disease Control and Prevention (CDC)
<http://www.cdc.gov>

Q. Is DEET safe?

A. Yes, products containing DEET are very safe when used according to the directions. Because DEET is so widely used, a great deal of testing has been done. When manufacturers seek registration with the U.S. Environmental Protection Agency (EPA) for products such as DEET, laboratory testing regarding both short-term and long-term health effects must be carried out. Over the long history of DEET use, very few confirmed incidents of toxic reactions to DEET have occurred when the product is used properly.

Insect Repellent Use

Q. Why should I use insect repellent?

A. Insect repellents help people reduce their exposure to mosquito bites that may carry potentially serious viruses such as West Nile virus, and allow them to continue to play and work outdoors.

Q. When should I use mosquito repellent?

A. Apply repellent when you are going to be outdoors and will be at risk for getting bitten by mosquitoes.

Q. What time of day should I wear mosquito repellent?

A. Many of the mosquitoes that carry the West Nile virus are especially likely to bite around dusk and dawn. If you are outdoors around these times of the day, it is important to apply repellent. In many parts of the country, there are mosquitoes that also bite during the day, and these mosquitoes have also been found to carry the West Nile virus. The safest decision is to apply repellent whenever you are outdoors.

Q. How often should repellent be reapplied?

A. Follow the directions on the product you are using in order to determine how frequently you need to reapply repellent. Sweating, perspiration or getting wet may mean that you need to re-apply repellent more frequently. If you are not being bitten, it is not necessary to re-apply repellent. Repellents containing a higher concentration of active ingredient (such as DEET) provide longer-lasting protection.

Q. Should I wear repellent while I am indoors?

A. Probably not. If mosquitoes are biting you while you are indoors, there are probably better ways to prevent these bites instead of wearing repellent all the time. Check window and door screens for holes that may be allowing mosquitoes inside. If your house or apartment does not have screens, a quick solution may be to staple or tack screening (available from a hardware store) across the windows. In some areas community programs can help older citizens or others who need assistance.

Q. How does mosquito repellent work?

A. Female mosquitoes bite people and animals because they need the protein found in blood to help develop their eggs. Mosquitoes are attracted to people by skin odors and carbon dioxide

from breath. Many repellents contain a chemical, N,N-diethyl-m-toluamide (DEET), which repels the mosquito, making the person unattractive for feeding. DEET does not kill mosquitoes; it just makes them unable to locate us. Repellents are effective only at short distances from the treated surface, so you may still see mosquitoes flying nearby. As long as you are not getting bitten, there is no reason to apply more DEET.

Q. Which mosquito repellent works the best?

A. The most effective repellents contain DEET (N,N-diethyl-m-toluamide), which is an ingredient used to repel pests like mosquitoes and ticks. DEET has been tested against a variety of biting insects and has been shown to be very effective. The more DEET a repellent contains the longer time it can protect you from mosquito bites. A higher percentage of DEET in a repellent does not mean that your protection is better—just that it will last longer. DEET concentrations higher than 50% do not increase the length of protection.

Q. How does the percentage of DEET in a product relate to the amount of protection it gives?

A. Based on a recent study:

- A product containing 23.8% DEET provided an average of 5 hours of protection from mosquito bites.
- A product containing 20% DEET provided almost 4 hours of protection
- A product with 6.65% DEET provided almost 2 hours of protection
- Products with 4.75% DEET and 2% soybean oil were both able to provide roughly 1 and a half hour of protection.

Choose a repellent that provides protection for the amount of time that you will be outdoors. A higher percentage of DEET should be used if you will be outdoors for several hours while a lower percentage of DEET can be used if time outdoors will be limited. You can also re-apply a product if you are outdoors for a longer time than expected and start to be bitten by mosquitoes.

Q. Why does CDC recommend using DEET?

A. DEET is the most effective and best-studied insect repellent available. (Fradin, 1998). Studies using humans and mosquitoes report that only products containing DEET offer long-lasting protection after a single application

Q. Are non-DEET repellents effective (e.g. Skin-So-Soft, plant-based repellents)?

A. Some non-DEET repellent products which are intended to be applied directly to skin also provide some protection from mosquito bites. However, studies have suggested that other products do not offer the same level of protection, or that protection does not last as long as products containing DEET. A soybean-oil-based product has been shown to provide protection for a period of time similar to a product with a low concentration of DEET (4.75%).

People should choose a repellent that they will be likely to use consistently and that will provide sufficient protection for the amount of time that they will be spending outdoors. Product labels often indicate the length of time that protection that can be expected from a product. Persons who are concerned about using DEET may wish to consult their health care provider for advice. The National Pesticide Information Center (NPIC) can also provide information through a toll-free number, 1-800-858-7378 or npic.orst.edu.

Q. I'm confused. Which products contain "DEET"?

A. Most insect repellents that are available in stores are labeled with the chemical name for DEET. Look for N,N-diethyl-m-toluamide or, sometimes, N,N-diethyl-3-methylbenamide. Choose a repellent that offers appropriate protection for the amount of time you will be outdoors. A higher percentage of DEET should be used if you will be outdoors for several hours while a lower percentage of DEET can be used if time outdoors will be limited.

Using Repellents Safely

Q. What are some general considerations to remember in order to use products containing DEET safely?

A. Always follow the recommendations appearing on the product label.

- Use enough repellent to cover exposed skin or clothing. Don't apply repellent to skin that is under clothing. Heavy application is not necessary to achieve protection.
- Do not apply repellent to cuts, wounds, or irritated skin.
- After returning indoors, wash treated skin with soap and water.
- Do not spray aerosol or pump products in enclosed areas.
- Do not apply aerosol or pump products directly to your face. Spray your hands and then rub them carefully over the face, avoiding eyes and mouth.

Q. How should products containing DEET be used on children?

A. No definitive studies exist in the scientific literature about what concentration of DEET is safe for children. No serious illness has been linked to the use of DEET in children when used according to the product recommendations. The [American Academy of Pediatrics](#) (AAP) Committee on Environmental Health has recently updated their recommendation for use of DEET products on children, citing: "Insect repellents containing DEET (N,N-diethyl-m-toluamide, also known as N,N-diethyl-3-methylbenzamide) with a concentration of 10% appear to be as safe as products with a concentration of 30% when used according to the directions on the product labels."

The AAP and other experts suggest that it is acceptable to apply repellent with low concentrations of DEET to infants over 2 months old. Other guidelines cite that it is acceptable to use repellents containing DEET on children over 2 years of age.

Repellent products that do not contain DEET are not likely to offer the same degree of protection from mosquito bites as products containing DEET. Non-DEET repellents have not necessarily been as thoroughly studied as DEET, and may not be safer for use on children.

Parents should choose the type and concentration of repellent to be used by taking into account the amount of time that a child will be outdoors, exposure to mosquitoes, and the risk of mosquito-transmitted disease in the area. Persons who are concerned about using DEET or other products on children may wish to consult their health care provider for advice. The National Pesticide Information Center (NPIC) can also provide information through a toll-free number, 1-800-858-7378 or npic.orst.edu.

Always follow the recommendations appearing on the product label when using repellent.

- When using repellent on a child, apply it to your own hands and then rub them on your child. Avoid children's eyes and mouth and use it sparingly around their ears.
- Do not apply repellent to children's hands. (Children may tend to put their hands in their mouths.)
- Do not allow young children to apply insect repellent to themselves; have an adult do it for them. Keep repellents out of reach of children.
- Do not apply repellent to skin under clothing. If repellent is applied to clothing, wash treated clothing before wearing again.

Using repellents on the skin is not the only way to avoid mosquito bites. Children and adults can wear clothing with long pants and long sleeves while outdoors. DEET or other repellents such as permethrin can also be applied to clothing (don't use permethrin on skin), as mosquitoes may bite through thin fabric. Mosquito netting can be used over infant carriers. Finally, it may be possible to reduce the number of mosquitoes in the area by getting rid of containers with standing water that provide breeding places for the mosquitoes.

Q. Is DEET safe for pregnant or nursing women?

A. There are no reported adverse events following use of repellents containing DEET in pregnant or breastfeeding women.

Q. Are there any risks due to using repellents containing DEET?

A. Use of these products may cause skin reactions in rare cases. If you suspect a reaction to this product, discontinue use, wash the treated skin, and call your local poison control center. There is a new national number to reach a Poison Control Center near you: 1-800-222-1222.

If you go to a doctor, take the product with you. Cases of serious reactions to products containing DEET have been related to misuse of the product, such as swallowing, using over broken skin, and using for multiple days without washing skin in between use, for example. Always follow the instructions on the product label.

Insect Repellents and Sunscreen

Q. Can I use an insect repellent containing DEET and sunscreen at the same time?

A. Yes. People can and should use both sunscreen and DEET when they are outdoors to protect their health. Follow the instructions on the package for proper application of each product. Apply sunscreen first, followed by repellent containing DEET.

To protect from sun exposure and insect bites, you can also wear long sleeves and long pants. You can also apply insect repellent containing DEET or permethrin to your clothing, rather than directly to your skin.

Q. Has CDC changed its recommendations for use of DEET and sunscreen?

A. No. Based on available research, CDC believes it is safe to use both products at the same time. Follow the instructions on the package for proper application of each product. Apply sunscreen first, then insect repellent containing DEET, to be sure that each product works as specified.

Q. Should I use a combination sunscreen/DEET-based insect repellent?

A. Because the instructions for safe use of DEET and safe use of sunscreen are different, CDC does not recommend using products that combine DEET with sunscreen.

In most situations, DEET does not need to be reapplied as frequently as sunscreen. DEET is very safe when applied correctly. The rare adverse reactions to DEET have generally occurred in situations where people do not follow the product instructions. Sunscreen often requires frequent reapplication, so using a combined product is not recommended. You do not need to reapply insect repellent every time you reapply sunscreen. Follow the instructions on the package for each product to get the best results.

Q. I heard about a study saying that there may be some type of interaction between repellents containing DEET and sunscreen. Is this true?

A. There has been attention to a study concerning the chemicals in DEET and sunscreen presented at a scientific meeting. This is an in vitro study, which means that it is a laboratory study that did not include human or animal testing. The goal of the study was to examine absorption of these chemicals, and it did not evaluate or make conclusions about health effects related to this issue. The study authors stated that further evaluation of the interaction of these chemicals should be conducted. The study has not yet been published (as of July 2003).

Evaluation by the EPA, which regulates products such as DEET, indicates that it is safe to use insect repellents containing DEET and sunscreen at the same time. CDC recommends using two separate products because sunscreen requires frequent applications while DEET should be used sparingly. Follow the directions on the package for each product, and consult your physician or pharmacist if you have questions. CDC's recommendations for the safe use of insect repellents on children and adults remain unchanged.

More information

Q. Where can I get more information about repellents?

A. For more information about using repellents safely please consult the EPA Web site: <http://www.epa.gov/pesticides/citizens/insectrp.htm> or consult the National Pesticide Information Center (NPIC), which is cooperatively sponsored by Oregon State University and the U.S. EPA. NPIC can be reached at: npic.orst.edu or 1-800-858-7378.