West Nile Virus: An Emerging Virus in North America

MELISSA NEDRY, CONNIE R. MAHON

West Nile virus is an emerging virus that first appeared in North America during the summer of 1999 in New York City. There were seven deaths associated with this event. Surveillance reports indicate that the virus had been spreading south and west and in 2002, had been reported in 42 states and the District of Columbia. As of September 2002, there were 2121 total human cases reported, including 104 deaths. The fatality rate for the West Nile virus is very low and the majority of individuals will have no clinical symptoms; however, individuals at most risk for more serious form of the disease are the elderly, the immunocompromised, and young individuals. The virus is spread by certain mosquito species and certain populations of birds serve as the reservoir hosts. Because person-to-person transmission does not occur, humans are therefore considered dead-end hosts. Confirmation of cases West Nile virus infections in humans are determined based on clinical and laboratory findings.

ABBREVIATIONS: CDC = Centers for Disease and Control; CSF = cerebrospinal fluid; EEE = Eastern Equine Encephalomyelitis; ELISA = enzyme-linked-immunosorbent assay; PCR = Polymerase Chain Reaction; PRNT = plaque reduction neutralization test; VEE = Venezuelan equine encephalitis; SLE = St Louis encephalitis; WNV = West Nile virus

INDEX TERMS: West Nile virus, encephalitis

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Melissa Nedry CLS *is a Clinical Laboratory Scientist at University Medical Center, San Antonio TX.*

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Connie R Mahon MS CLS *is Director, Clinical Laboratory Science Program, Walter Reed Army Medical Center, Washington DC.*

Address for correspondence: *Connie R Mahon MS CLS, Director, Clinical Laboratory Science Program, Walter Reed Army Medical Center, Department of Pathology and Area Laboratory Services (DPALS), 6900 Georgia Ave NW, Washington DC 20307-5002, (202) 782-8353, (202) 782-4502 (fax). connie.mahon@na.amedd.army.mil*

Connie R Mahon MS CLS *is the Focus: Microbiology guest editor.*

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LEARNING OBJECTIVES

1. Describe the general viral characteristics of West Nile virus.
2. Name the species of the primary vectors that have been identified to transmit West Nile virus.
3. List three reservoir hosts for West Nile virus.
4. Identify the two populations at highest risk for encephalitis from West Nile virus infections.
5. Describe four laboratory confirmation criteria established by CDC to identify West Nile virus.

West Nile virus (WNV) was first isolated in 1937 from the blood of an ill woman in the West Nile region of Uganda. The original investigators noted at the time that the virus caused encephalitis in rhesus monkeys. During the next 15 years, the disease was found to be endemic in Egypt and had caused sporadic summertime epidemics in Israel. In the 1950s, attack rates in Israel were at times greater than 60%. Since, infection has been found in Africa, the Middle East, parts of Europe, the Indian subcontinent, and the former Soviet Union.[1] In 1974, the largest human outbreak of West Nile encephalitis occurred in South Africa and in 2000, a huge outbreak occurred in Israel with 417 reported cases.[2]

The first WNV encephalitis outbreak in the Western Hemisphere occurred in the late summer and fall of 1999 in New York state; 62 cases were reported, seven fatal cases occurred in New York City (NYC) and two in other counties. In 2000,

Table 1. West Nile virus current case count*

State	Laboratory positive human cases	Deaths
Alabama	25	1
Arkansas	11	1
California	1	—
Colorado	1	—
Connecticut	7	—
District of Columbia	6	—
Florida	8	—
Georgia	19	5
Illinois	518	29
Indiana	104	—
Iowa	18	—
Kentucky	27	4
Louisiana	261	11
Maryland	6	—
Massachusetts	10	2
Michigan	270	13
Minnesota	19	—
Mississippi	157	6
Missouri	114	3
Nebraska	48	3
New Jersey	4	—
New York	46	3
North Carolina	1	—
North Dakota	15	2
Ohio	232	9
Oklahoma	4	—
Pennsylvania	18	3
South Carolina	1	—
South Dakota	23	—
Tennessee	26	4
Texas	91	2
Virginia	16	1
Wisconsin	14	2
Totals	2121	104

* As of September 25, 2002 these are the human case totals for 2002 that have been reported to CDC/Arbonet or compiled in direct communication with state and local health officials. Arbonet is the national, electronic surveillance system established by CDC to assist states in tracking West Nile and other mosquito-borne viruses.

Source: Centers for Disease Control and Prevention. Office of Communication. West Nile Virus Update: Current Case Count. http://www.cdc.gov/od/oc/media/wncount.htm. Accessed September 25, 2002.

21 cases were reported in New York, including two deaths in the NYC area.[3] In 2001, 66 human cases of WNV encephalitis or meningitis were reported from 39 counties in ten states. Nine cases were fatal.[4] As of September 25, 2002, the human case totals for 2002 that have been reported to the Centers for Disease Control and Prevention (CDC) or compiled in direct communication with state and local health officials were 2,121 including 104 deaths. Table 1 shows the latest WNV case count (as of September 25, 2002).[5] Laboratory confirmed human cases were reported in 33 states. Illinois has reported the most number of cases (518) with 29 fatalities.[5]

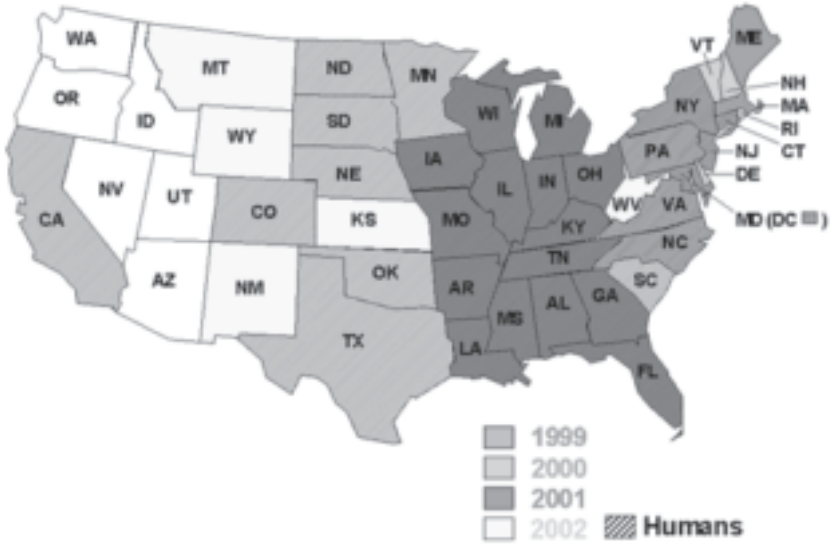
Until recently, outbreaks of WNV infections in humans occurred infrequently. However, since the mid-1990s, epidemiologic trends for WNV have been noted and have caused serious concerns: 1) frequency of outbreaks in humans and horses has increased, 2) increased severity of human disease, and 3) high avian death rates during human outbreaks, particularly in outbreaks in Israel and the U.S. It is still unknown whether high avian death rates in the U.S. are caused by higher virulence of the circulating WNV strains or increased susceptibility of North American birds. Nevertheless, high avian death rates during the 1999 epizootic in the NYC area incited an avian mortality surveillance to determine the spread of WNV in the eastern and southern U.S.

In 1999, state and local health departments in the eastern United States in collaboration with CDC instituted surveillance systems to detect WNV human infections. ArboNET, a Web-based, national surveillance data network established by the CDC is maintained by 54 state and local public health agencies. The surveillance systems aim to promote timely detection and reporting of meningoencephalitis occurrences and assist states in tracking West Nile and other mosquito-borne viruses.[2]

In 2000, WNV surveillance showed WNV infection in birds in 12 states and the District of Columbia. The report showed a total of 4,139 WNV-infected dead birds found in 133 counties and in which crows were the most frequently reported WNV-infected species.[6,7] In 2001, a marked increase was noted when the WNV surveillance data reported activity in 359 counties in 27 states. Infected horses, the only nonhuman mammal infected with the virus, were reported in 14 states.[4,8] However, as of September 2002, 4,562 dead crows and 3,366 other dead birds with WNV have been reported from 42 states, NYC, and the District of Columbia; 2,244 WNV infections in mammals (includes all equine) have been reported from 31 states.[9]

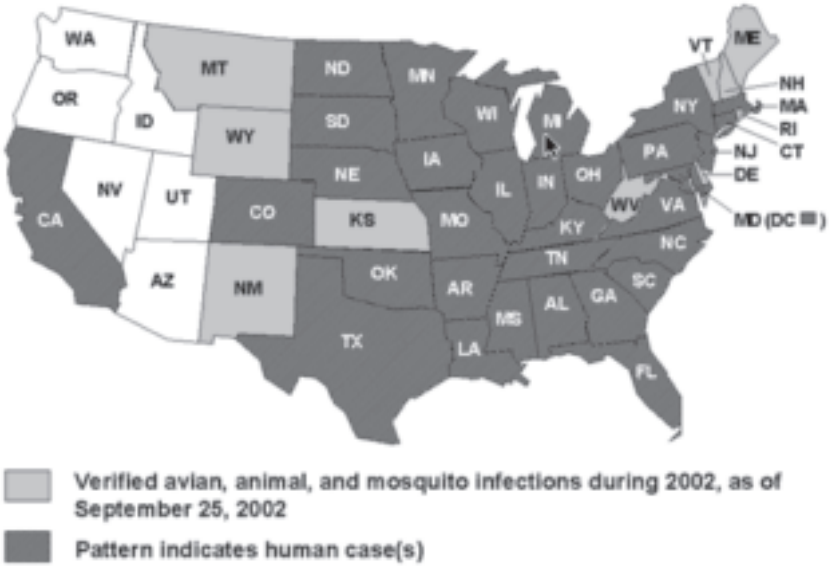
Avian mortality surveillance in 2000 reported geographic expansion of WNV activity although human infections were reported only in NYC and surrounding counties in New Jersey and Connecticut. Ten of the 21 human cases identified in 2000 lived on Staten Island, the only county in NYC where WNV in humans was not found in 1999. The cause for the focal human epidemic in 2000 despite the geographically expanding epizootic remains unknown.[3,7]

Figure 1. West Nile Virus in the United States, 1999-2002



Source: Centers for Disease Control and Prevention. Statistics, Surveillance, and Control. http://www.cdc.gove/ncidod/dvbid/westnile/srv&control.htm Accessed 25 September 2002

Figure 2. West Nile Virus in the United States, 2002



Source: Centers for Disease Control and Prevention. Statistics, Surveillance, and Control. http://www.cdc.gove/ncidod/dvbid/westnile/srv&control.htm Accessed 25 September 2002

Nevertheless, the 2001 surveillance indicate an increase geographically in WNV activity. Figures 1 and 2 show the spreading WNV activity in the US from 1999–2002.[10] The figures also show where human cases have been reported.[10] The 2001 surveillance findings have also established the notable spread of WNV westward and southward. This report shows the concurrent appearance of two epizootic foci: one in the mid–Atlantic region and the other in the southeast along the shared borders of Florida, Georgia, and Alabama—which suggests that migratory birds introduced WNV into the southeastern states. Migratory bird species use fixed north-south flyways; hence, movement of WNV from the mid-Atlantic region to the south-Atlantic region and the Gulf states was anticipated. Causes for the WNV spread into multiple foci in the central U.S. however, are not as apparent. The return of WNV infected birds from the south or their travels from east-to-west has been considered as a likely source.[4]

THE VIRUS

WNV, an RNA virus, belongs to the Fflavivirus family that consists of over 70 viruses. Flavivirus family includes viruses such as Japanese encephalitis (JE), yellow fever, St Louis encephalitis, dengue, and tick-borne encephalitis, etc.[9] Flaviviruses measure between 40–60 nm; are enveloped, icosahedral nucleocapsid, nucleic acid (positive-sense, single stranded RNA, approximately 10,000 to 11,000 bases); and present a similar appearance in the elec-

tron microscope.[11] Because all flaviviruses are closely related antigenitically, serologic cross-reactions are not uncommonly observed in the diagnostic laboratory. The WNV was originally identified as St Louis encephalitis (SLE) virus because they share 80% homology.[12] Hence, WNV has been placed in the JE virus serocomplex, which includes viruses that have been associated with human encephalitis: JE, SLE, Murray Valley encephalitis, and Kunjin (a subtype of WNV). Viruses in the JE complex are so closely related that specialized tests are often required to determine the infecting flavivirus. Acute and convalescent serum samples from patients are necessary to fully evaluate the antibody response.[3]

WNV can be divided into two genetic lineages: Lineage 1, the lineage of the WNV that has only been associated with clinical human encephalitis and lineage 2, which, unlike lineage 1, has not been involved in any clinical human encephalitis. Lineage 1 WNV has been isolated from India, Africa, Europe, Asia, and North America while WNV lineage 2 has circulated in Africa. The WNV circulating in Israel is the closest relative to the New York strain from the NYC outbreak in the summer of 1999.[3]

EPIDEMIOLOGY AND TRANSMISSION

It probably will remain unknown how WNV was introduced when it first appeared in NYC in the summer of 1999. It is known, however, that the WNV isolates from New York appeared to be genetically related to the WNV isolates from Israel. This suggests that the virus may have originated from the Middle East. Investigators speculate that introduction may have occurred via: 1) infected persons traveling to New York, 2) importation of infected birds or mosquitoes, or 3) infected birds migrating to the continent.[3]

It is also known that transmission of the WNV occurs between the natural bird (reservoir hosts) and the vector (mosquito). The *Culex* family (*Culex pipiens*) and the *Aedes* family (*Aedes vexans*) are the primary mosquito vectors identified, although during 2000, nine other mosquito species were determined to carry WNV.[2] Currently, studies are under way to determine if the species are able to transmit the WNV by bite before they can be considered as vectors. Additional factors to be considered include host preference, feeding behavior, population density, longevity, and seasonal activity of each mosquito.[13]

WNV is carried in the salivary glands of the vector mosquito and infects birds during blood-meal feeding. Strong birds develop infectious viremia within one to four days after exposure and the reservoir hosts then develop life-long immunity. Since July 2001, more than 70 species of birds have tested positive for the WNV. Other viruses such as SLE and eastern equine encephalomyelitis (EEE) have a similar enzootic cycle involving birds as hosts and mosquitoes as vectors. Handling live or dead infected birds has not been shown to transmit WNV to humans and no complicated illness has been observed in animals such as cats and dogs. There is no documented evidence of person-to-person or animal-to-person transmission for WNV. However, the WNV has been recognized to have caused several deaths in horses. WNV infections in chipmunks, bats, raccoons, skunks, squirrels, and domestic rabbits have been reported to the CDC.[14]

The risk for WNV infection via donated blood or organs is not known because until recently, WNV infection in organ transplant recipients had not been reported. In August 2002, three of four organ transplant recipients from the same donor developed WNV meningoencephalitis and the fourth one, WNV fever. All four organs were recovered from a previously healthy individual who suffered a fatal injury. Before death, the organ donor received several transfusions of blood products. The donor serum collected before the organs were obtained was tested by polymerase chain reaction (PCR) and revealed the presence of WNV. The source of the organ donor's infection is still unknown and investigations of the transfusions the organ donor received continue.[15]

PATHOGENESIS AND CLINICAL MANIFESTATIONS

The majority of WNV infections are clinically silent. Although individuals infected with the WNV have less than 1% chance of developing severe illness, the fatality rate ranges from 3% to 15% with the elderly being at highest risk.[16] The elderly and young adolescents are also at the highest risk of developing severe encephalitis.[17] The incubation period ranges from two to six days. In mild infections, fever, periocular pain, lymphadenopathy, malaise, muscle pain, and gastrointestinal symptoms, which last about three to five days, appear. A biphasic pattern may be seen with the fever similar to dengue fever (acute illness for a few days, spur-of-the-moment remission, again recurrence). Meningitis or encephalitis develops in less than 1% of infected individuals. Symptoms include stiff neck, high fever, headache, muscle weakness, disorientation, tremors, convulsions, and paralysis. In encephalitis, changed mental status or other cortical signs are seen while patients with meningoencephalitis present with neurological manifestations common to both conditions.[18]

Because clinical symptoms resemble a mild viral illness, the diagnosis of WNV infections is often difficult to establish. During the NYC summer 1999 outbreak, the human

cases were first thought to be caused by the SLE virus because laboratory, clinical, and epidemiologic data correlated well with the SLE.[19] Although SLE infection most often displays more cerebral involvement than WNV, and unlike the WNV, more fulminant changes are seen pathologically in the Eastern and Western equine encephalitis virus infections, differentiating WNV infections from those caused by viruses in the flavivirus family remains a challenge clinically and in the laboratory.[18]

A polio-like syndrome, acute flaccid paralysis (AFP), has also been described in six patients with acute WNV infection. All six patients presented with an acute onset of painless asymmetrical weakness and without sensory loss. Although the exact cause of AFP in these patients has not been fully evaluated, clinical and electrophysical findings so far suggest a disease process similar to that seen in acute poliomyelitis. Patients who present with AFP, which is attributed to peripheral demyelination as in Guillain-Barre´ syndrome, must be evaluated for indications of WNV infection. WNV infection-associated AFP must be differentiated from other forms of acute flaccid paralysis such as Guillain-Barre´ syndrome and acute poliomyelitis.[20]

LABORATORY DIAGNOSIS

During the outbreaks in the New York area in 1999 and 2000, WNV human infections were confirmed based on the patient's clinical presentation and laboratory test results. Clinical findings included hospitalization with an illness associated with central nervous system manifestations consistent with meningitis or encephalitis while laboratory diagnosis was based on the four laboratory confirmation criteria established by CDC: 1) recovery of WNV from tissue or demonstration of viral particles or genomic sequences in tissue; 2) detection of IgM to WNV in CSF by IgM-capture ELISA; 3) >4-fold serial change in plaque-reduction neutralizing antibody titer (PRNT) to WNV in paired sera or CSF samples; and 4) demonstration of both WNV-specific IgM (by ELISA) and IgG (ELISA screened and PRNT confirmed) antibody in a single serum sample.[18]

There are currently no commercial kits available to detect WNV infections serologically. Most requests for WNV testing are sent to local state health department laboratories for initial screening for the presence of IgM antibody in blood or CSF. Enzyme-linked immuno-sorbent assay (ELISA) has been used as a serologic screening test for the WNV infection. IgM

Table 2. West Nile patient laboratory findings, New York and New Jersey, 1999 and 2000

Test	Number tested (%)	Mean value (range)	Normal values[2]
CSF			
Leukocyte count, mean	19 (100)	308 x 10^6/L (0-1782)	0-5 cells x 10^6/L
Red cell count, mean	16 (84)	115 x 10^6/L (0-700)	0 cells x 10^6/L
Protein, mean	19 (100)	111 mg/dL (56-555)	15-50 mg/dL
Glucose, mean	19 (100)	67 mg/dL (48-95)	50-80 mg/dL
Differential,* ≥50% neutrophils	15 (79)	9 (1-100%)	All mononuclear cells
Complete blood cell count			
Leukocyte count, mean	19 (100)	10.6 x 10^9/L (4.4-19.7)	4.5-11.0 x 10^9/L
Differential cell count,*>77% segs + bands	18 (95)	11 (55-96%)	59% ± 18
Hemoglobin (male), mean	11 (100)	14.5 g/dL (11.8-16.5)	15.5 g/dL ± 1.1
Hemoglobin (female), mean	8 (100)	12.7 g/dL (10.5-14.6)	13.7 g/dL ± 1.0
Other laboratory		**N with condition (%)**	
Hyponatremia, serum Na <135 mmol/L	19(100)	8 (42%)	135-145 mmol/L
Elevated AST, >twice upper limit	17 (90)	4 (24%)	10-35 units/L
Elevated ALT, >twice upper limit	15 (79)	1 (7%)	20-48 units/L
Elevated total bilirubin, >twice upper limit	16 (84)	3 (19%)	0.3-1.0 mg/dL

*Values are the number of patients with the laboratory finding; ranges are the values of all patients. CSF = cerebrospinal fluid; AST = aspartate aminotransferase; ALT = alanine aminotransferase; segs = segmented neutrophils.

Source: Weiss D, Carr D, Kellachan J, and others. Clinical findings of West Nile virus infection in hospitalized patients, New York and New Jersey, 2000. Emerg Infect Dis 2001;7(4).

and IgG ELISA antibody tests are performed to establish whether the patient has a current infection or has had a previous exposure. If the patient has been immunized for yellow fever or JE, or if the patient is infected with SLE, dengue, or other flavivirus, false-positive results for the WNV infection may occur. If a patient shows only IgM antibody in a single serum, the individual may be classified as someone having a probable recent infection. However, if the patient shows IgG antibody, a more specific plaque reduction neutralization test (PRNT) should be performed. Paired samples of acute-phase and convalescent-phase serum samples are therefore more helpful for demonstration of seroconversion than single serum collection. A positive WNV infection shows a ≥4-fold serial change in the PRNT in serum or cerebrospinal fluid (CSF) samples.[21] IgM antibody to WNV is also detected in the CSF of patients who have encephalitis as early as the first few days of illness by antibody capture-ELISA. Polymerase chain reaction (PCR) is used for the detection of viral nucleic acid from human-tissue specimens.[2] Other laboratory findings for WNV infections are shown on Table 2.[18]

TREATMENT AND PREVENTION

The appearance of WNV in the U.S. during the summer and fall of 1999 caused major concerns regarding the preparedness of public health agencies in handling vector-borne diseases during sporadic and outbreak events.[18] Until recently, because of the sporadic nature of the disease in other continents, development of the WNV vaccine for animals and humans has not been considered a priority. Research priorities now include WNV vaccine development for animals and humans and although a WNV vaccine for horses has been licensed recently, its efficacy remains to be determined.[14] Similarly, antiviral therapy for WNV infection has not been established although Ribavirin has shown to inhibit the WNV replication and cytopathic effects in neural cell cultures.[17,22,23] The drug is a very-broad spectrum virustatic antiviral agent that has multiple mechanisms of action. As a result, viral resistance rarely develops and it can be administered orally, intravenously, or via a nebulizer. Degrees of efficacy have been shown in a variety of human viral diseases.[23]

PREVENTIVE MEASURES

Mosquito-control has been described to be the most effective means to prevent WNV transmission. Hence, the United States Environmental Protection Agency (EPA) has established several precautionary measures to follow.[23,24]

- Empty standing water in old tires, cemetery urns, buckets, plastic covers, toys, or any other container where "wrigglers" and "tumblers" live.
- Empty and change the water in bird baths, fountains, wading pools, rain barrels, and potted plant trays at least once a week, if not more often.
- Drain or fill temporary pools with dirt.
- Keep swimming pools treated and circulating and rain gutters unclogged.
- Use mosquito repellents when necessary and follow label directions and precautions closely. The American Chemical Society has presented new research for mosquito repellents such as the oil in catnip which gives a distinctive minty odor to repel mosquitoes. More research is needed however to determine the effects on humans.[25]
- Use head nets, long sleeves, and long pants if you venture into areas with high mosquito populations, such as salt marshes.
- If there is a mosquito-borne disease warning in effect, stay inside during the evening when mosquitoes are most active.
- Make sure window and door screens are "bug tight."
- Replace your outdoor lights with yellow "bug" lights.
- Contact your local mosquito control district or health department.

Neighborhoods are occasionally sprayed to prevent disease and nuisance caused by large mosquito numbers. If you have any questions about mosquitoes and their control, call your local authorities.

CONCLUSION

WNV is an emerging virus that is continuously spreading throughout North America and is showing patterns of moving south and west in the U.S. As of early Fall 2002, more than two thousand human cases of WNV infections have been reported, including over 100 deaths. Although the majority of infected individuals will not show symptoms, the elderly, those who are immunocompromised, and young individuals are at most risk for the serious form of the disease. Common infection control methods should be practiced to prevent the spread of the infection. To learn more about WNV, sites such as CDC (www.cdc.gov), WNF (www.westnilefever.com) and EPA (www.epa.gov) can be accessed for the latest update and information.

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