West Nile virus infection

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Purpose. The epidemiology, virology, and transmission of West Nile virus (WNV) are reviewed, and the clinical features, diagnosis, and treatment of WNV infection are examined.

Summary. WNV infection is caused by a flavivirus transmitted from birds to humans through the bite of culicine mosquitoes. WNV was discovered in the blood of a febrile woman from Uganda's West Nile province in 1937. The first case of domestically acquired WNV infection was reported in the United States in 1999 in New York. Since then, WNV infection has spread rapidly across the United States, with 9306 confirmed cases and 210 deaths reported from 45 states in 2003. It is still not clear how WNV was introduced into North America. WNV is a small, single-stranded RNA virus and a member of the Japanese encephalitis virus antigenic complex. While most humans infected with WNV are asymptomatic, some may develop an influenza-like illness. Disease surveillance remains the cornerstone for the early recognition and control of WNV. We describe one case of WNV infection with an update on the disease. Strategies for the prevention and control of this infection are reviewed.

Conclusion. There is no established treatment for WNV infection. Currently, prevention and control are the only measures that help decrease the morbidity and mortality associated with WNV infection. As the number of cases escalates and the geographic distribution of WNV infection widens, the epidemic will continue to pose a major challenge to clinicians in the coming years. There is an urgent need for more research on the pathogenesis and treatment of WNV infection.

Index terms: Diagnosis; Epidemiology; Mortality; West Nile fever

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Within the past five years, West Nile virus (WNV), a mosquito-borne flavivirus and human neuropathogen commonly found in West Asia, Africa, and the Middle East, has emerged as an important human, avian, and equine disease in the United States.[1-8] Most people infected with WNV remain asymptomatic, and about 20% develop mild flu-like symptoms. About 1 (<1%) in 150 develop acute neurologic disease, which can lead to coma, stupor, paralysis, and death.[9,10]

Clinical case

A 55-year-old African-American man from upstate New York was admitted in August 2002 with a chief complaint of fever and confusion. According to the patient's wife, he had been healthy until one day before admission when he complained of flu-like symptoms, including nausea, vomiting, and diarrhea. The patient believed these symptoms were from eating contaminated venison sausage. His medical history was significant for hypertension and he denied the use of tobacco, alcohol, and illicit drugs. Other pertinent history includes no recent travel out of the area and previous employment in a factory. The patient was also an avid hunter. Physical examination revealed a well-developed, well-nourished individual who was confused and agitated. While most of the patient's physical examination findings were normal, his neck was rigid. Vital signs included a temperature of 39.9 °C, blood pressure of 120/70 mm Hg, a respiration rate of 22 breaths per minute, and tachycardia. On neurologic examination the patient showed signs of hyperreflexia of the upper and lower extremities and no evidence of focal signs of motor

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weakness. Both Babinski's sign and computed tomography of the brain were negative. Chest radiograph was also read as negative. Laboratory tests revealed a serum peripheral leukocyte count of $12 \times 10^{-3}/\mu L$, with 84% neutrophils and 16% lymphocytes. His serum creatinine phosphokinase level was 336 units/L. Cerebrospinal fluid (CSF) analysis revealed 62 white blood cells/μL, 51 red blood cells/μL, with 67% neutrophils, 18% lymphocytes, 12% monocytes, a glucose concentration of 72 mg/dL (serum glucose was 90 mg/dL), and protein levels of 51 mg/dL. Gram's stain, India ink capsule stain, and cryptococcal antigen capture assay of the CSF were negative.

Based on these findings, the patient's working diagnosis was herpes encephalitis, and i.v. acyclovir 800 mg was administered every 8 hours. The patient also received i.v. ampicillin 2 g every 4 hours and i.v. ceftriaxone 2 g every 12 hours with the intent of discontinuation on the return of negative bacterial cultures. After five days, the patient's fever resolved, and his mental status improved. The acyclovir was discontinued when the herpes polymerase chain reaction (PCR) was negative. Although initial WNV serology test results were negative, CSF WNV PCR and CSF immunoglobulin M (IgM) ELISA results were positive. Repeat WNV immunoglobulin G and IgM serology tests were positive two weeks later. Magnetic resonance imaging revealed a left basal ganglion lesion. The patient improved after three weeks but complained of persistent lower extremity weakness and was discharged to a rehabilitation facility. After two months, all of his symptoms resolved, and he was able to return to work.

Epidemiology

WNV was first discovered in the blood of a febrile woman from Uganda's West Nile province in 1937. Outbreaks in humans mainly associated with mild febrile illness were reported infrequently in Israel and Africa until the early 1990s.[11] However, outbreaks involving hundreds of people resulting in severe neurologic disease were reported in Romania, Russia, and Israel in the late 1990s.[12-14] Although a large outbreak of WNV has yet to occur in the United States, the frequency and territory occupied by this disease have been increasing since 1999.[15] It is unclear if changes in the severity and frequency of WNV infection are due to changes in host factors, such as age, background immunity, or predisposing chronic conditions, or to increasing virus virulence.[16]

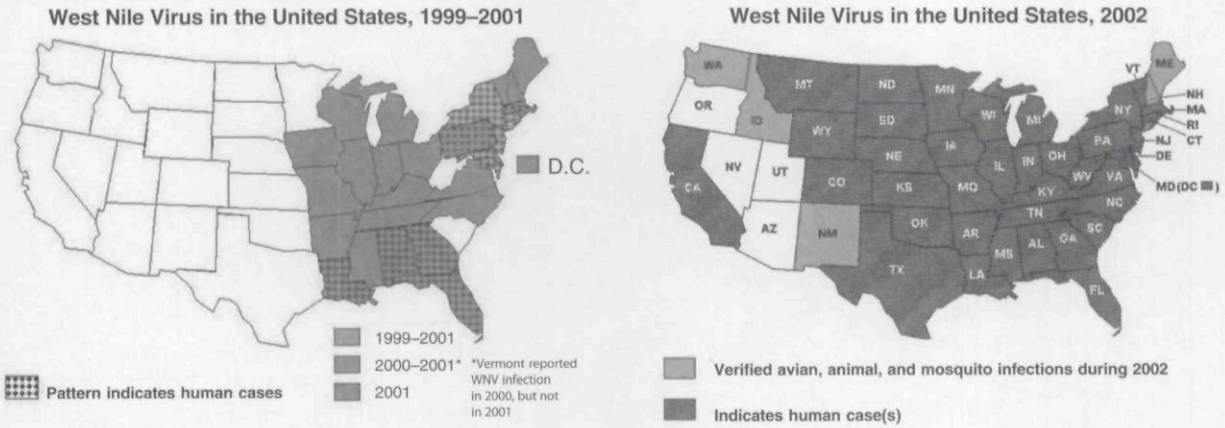
The first domestically acquired human cases of West Nile encephalitis were reported in Queens, New York, in the summer of 1999 and resulted in 59 clinical cases and 7 deaths; the median age of these patients was 71 years.[17] Since then, the disease has quickly spread to the Midwest, along the Eastern seaboard, and into the Deep South. In 2002, there were 4156 laboratory-confirmed human cases, including 2942 meningoencephalitis cases and 284 deaths (9% fatality rate).[18-21] From 1999 through October 22, 2003, WNV infection had been reported in 45 states (Figure 1).[22] As of October 22, 2003, 7386 human cases and 155 WNV-associated deaths were reported in 2003.[22] Fifty-three percent of the infected patients were men, and the median age was 47 years. In addition, 10,453 dead birds with WNV infection were reported from 42 states in 2003.[22] It is interesting to note that the first case of West Nile encephalitis was reported in New York in the early 1950s when the virus was unsuccessfully used as an experimental treatment for advanced cancer.[23] WNV is also spreading rapidly throughout Canada.[24] It was first discovered in 2001 in birds and mosquitoes in Ontario. Subsequent cases were reported in Nova Scotia, Quebec, Manitoba, Ontario, and Saskatchewan in 2002, with cases of infection reported in Quebec and Ontario in 2002. The peak transmission time for WNV ranges from mid-July to early December, most commonly in late August and early September.[16]

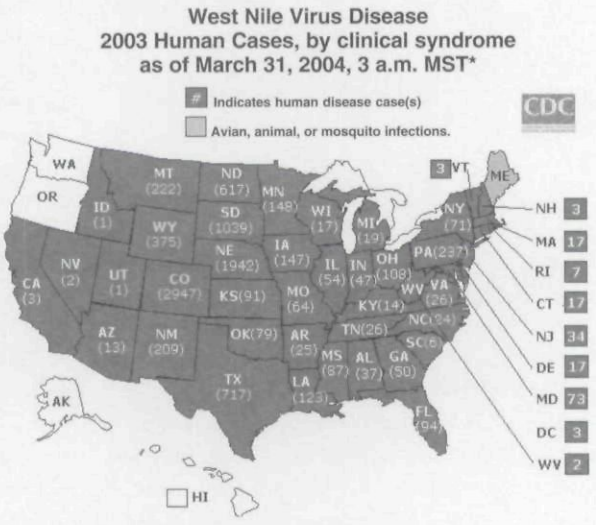
Virology

The WNV is a small, single-stranded RNA virus of the family Flaviviridae and genus *Flavivirus* and a member of the Japanese encephalitis virus antigenic complex.[17,25] The complex includes Alfuy, Cacipacore, Japanese encephalitis, Koutango, Kunjin, Murray Valley encephalitis, St. Louis encephalitis, Rocio, Stratforde, Usutu, Yaounde, and WNV. The flaviviruses measure 40–60 nm in diameter, are symmetrical in shape, and have positive-sense single-stranded RNA. As with other members of the Japanese encephalitis virus antigenic complex, WNV possesses E-glycoprotein as the viral hemagglutinin and mediator of virus–host cell binding.[11,16,25] E-glycoprotein elicits most of virus-neutralizing antibodies. The flavivirus virion has an icosahedral core composed of multiple copies of a capsid protein, which encloses positive-sense single-stranded RNA. A host–cell-derived envelope encloses the capsid. This close antigenic relationship between the flaviviruses accounts for the serologic cross-reactions observed in the laboratory.[16]

Although two genetic lineages of WNV have been identified, all North American strains are closely related to phylogenetic lineage I WNV.[11] Lineage II WNV remains in enzootic foci in Africa, where large outbreaks of encephalitis have not been reported.[26] It has been suggested that lineage II WNV may be less virulent in humans[27]; however, a mouse model has demonstrated that WNV from both lineages is neuroinvasive.[28] Although WNV has caused infections worldwide, mortality from WNV infection has occurred only in the United States and Israel.

Figure 1. Status with reported West Nile virus activity in the United States, 1999–2003. Available from www.cdc.gov/ncidod/dvbid/westnile/surv&control03Maps99_01.htm,www.cdc.gov/ncidod/dvbid/westnile/surv&control03Maps02.htm, and www.cdc.gov/ncidod/dvbid/westnile/surv&control03Maps.htm.





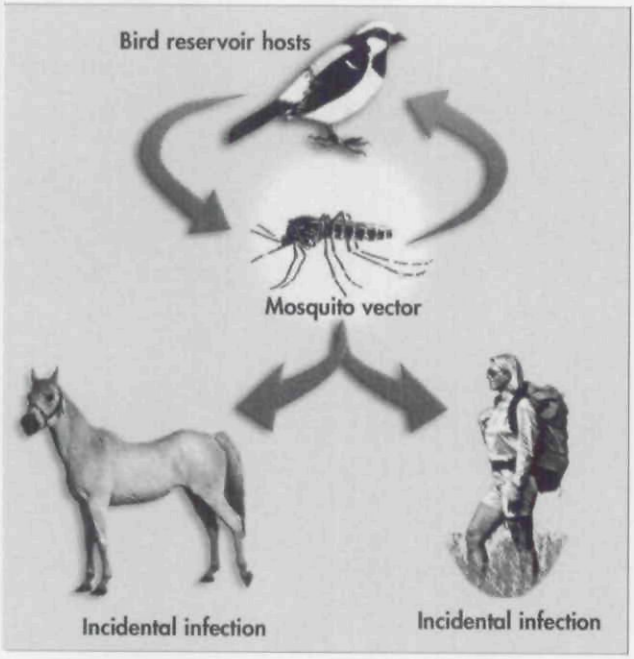
Ecology and transmission

It is still not clear how WNV was introduced into North America. Possible mechanisms include importation or migration of infected birds or mosquitoes and international travel of infected persons.

WNV is maintained in a enzootic cycle involving several species of mosquitoes and birds before infecting humans (Figure 2). Hence, the most common route of WNV transmission to humans is through the bite of an infected mosquito.[29,30] Mosquitoes become infected with WNV when they feed on a bird carrying the virus in its blood. Birds in the Corvidae family (e.g., crows, blue jays) are particularly susceptible vectors because of their high viremia.[23] While it is unclear which mosquito species is primarily responsible for WNV transmission to humans, *Culex pipiens, Culex restuans,* and *Culex quinquefasciatus* appear to be the virus's most important vectors. WNV transmission occurs between birds and maintenance-vector mosquitoes until the early fall. Twenty-seven mammalian species have been shown to be susceptible to WNV infection and disease, including humans and horses.[29,30] Within 10–14 days of becoming infected, a mosquito can transmit the virus in its saliva to another bird or animal. During the bird–mosquito–bird cycle, viral amplification occurs, increasing the likelihood of WNV infection when an individual is bitten.[29,30]

In 2002, four novel routes of WNV transmission to humans were reported: (1) blood transfusion, (2) organ transplantation, (3) transplacental transfer, and (4) breastfeeding.[5,7,8,31-33] In 2002, 23 people in the United States were reported to

Figure 2. West Nile virus transmission cycle. Reprinted from reference 30, with permission.



have acquired WNV infection after receiving blood components from 16 WNV-infected blood donors.[7] Per the recommendation of the Food and Drug Administration, blood-collection agencies have since implemented WNV nucleic acid amplification tests to screen all blood donations in order to quarantine infectious blood components.[8] The implementation of national blood-donor screening has significantly reduced the risk of WNV transmission by removing hundreds of units of potentially infectious blood donated by asymptomatic donors. WNV titers in infected blood components can be as low as 0.8 plaque-forming units/mL, which is lower than titers reported for other blood-borne viral pathogens, such as human immunodeficiency virus and hepatitis C virus.[7] As of September 16, 2003, 489 WNV-viremic blood donors and two cases of confirmed transfusion-associated events had been reported, demonstrating that the risk of transmission has not been eliminated by screening, possibly because of low viremia or other unknown mechanisms.[8]

Clinical features

While most humans infected with WNV are asymptomatic, some may develop an influenza-like illness, manifested by sudden onset of high fever with chills after an incubation period of 3–14 days, and symptoms usually last 3–6 days.[34,35] Patients may also have malaise, headache, arthralgia, lymphadenopathy, backache, anorexia, vomiting, myalgia, and retro-orbital eye pain made worse by eye movement.[34,35]

Of more concern is West Nile encephalitis which was first documented in 1957 in an Israeli nursing home.[16] While more recent outbreaks of West Nile encephalitis have been associated with morbidity and mortality, reports of severe neurologic disease caused by WNV remain uncommon.[16,34] WNV-related neurologic disease can manifest as meningitis, encephalitis, or acute flaccid paralysis (Table 1).[9,10,12] More than 90% of patients with neurologic disease have fever accompanied by gastrointestinal symptoms and headache.[10] Meningoencephalitis has been reported more frequently than meningitis alone in patients with West Nile encephalitis.[12,14] WNV may also cause a flaccid paralysis of the limbs and respiratory tract, as seen in many patients during the 1999 outbreak in New York City.[17] Other neurologic symptoms, such as tremor, myoclonus, and parkinsonian features (including rigidity, bradykinesia, and postural instability) have been reported with WNV infection.[10,36,37]

Age is an important prognostic indicator, as younger patients are more likely to develop West Nile fever alone. On the other hand, the risk of developing severe neurologic disease increases dramatically in patients over 50 years of age.

Diagnosis

Indirect immunofluorescence assays using murine monoclonal antibodies are sensitive for rapid testing of mosquito pools, avian tissues, and human specimens.[35,38,39] WNV infection should always be considered in patients with encephalitis and meningitis whose cause cannot be determined. This is especially true in elderly patients with encephalitis who develop these illnesses in the late summer or early fall. Detection of IgM antibody to WNV in serum or CSF using ELISA is the most efficient diagnostic method. As IgM does not cross the blood–brain barrier, IgM antibody to WNV detected in the CSF strongly suggests central nervous system involvement.[34]

In most patients, IgG antibodies can be detected three weeks after infection. IgG titers increase between days 7 and 21 in patients with acute infection. Clinicians should be aware that false-positive serologic results can occur in patients recently vaccinated against yellow fever or Japa-

Table 1.
Neurologic Manifestations of West Nile Virus[a]

Diagnosis	Criteria
West Nile meningitis	A. Clinical signs of meningeal inflammation, including nuchal rigidity, Kernig or Brudzinski sign, or photophobia or phonophobia B. Additional evidence of acute infection, including one or more of the following: fever (>38 °C) or hypothermia (<35 °C); cerebrospinal fluid pleocytosis (≥5 leukocytes/mm^3); peripheral leukocyte count >10,000/mm^3; neuroimaging findings consistent with acute meningeal inflammation
West Nile encephalitis	A. Encephalopathy (altered level of consciousness, lethargy, or personality change lasting ≥24 hr) B. Additional evidence of central nervous system inflammation, including two or more of the following: fever (≥38 °C) or hypothermia (≤35 °C); cerebrospinal fluid pleocytosis (≥5 leukocytes/mm^3); peripheral leukocyte count >10,000/mm^3; neuroimaging findings consistent with acute inflammation (with or without involvement of the meninges) or acute demyelination; presence of focal neurologic deficit; meningisimus (as defined in A); electroencephalography findings consistent with encephalitis; new onset or exacerbation of previously controlled seizures
Acute flaccid paralysis	A. Acute onset of limb weakness with marked progression over 48 hr B. At least two of the following: asymmetry to weakness; areflexia/hyporeflexia of affected limb(s); absence of pain, paresthesia, or numbness in affected limb(s); cerebrospinal fluid pleocytosis (≥5 leukocytes/mm^3) and elevated protein levels (≥45 mg/dL); electrodiagnostic studies consistent with an anterior horn cell process; spinal cord magnetic resonance imaging documenting abnormal increased signal in the anterior gray matter

[a]Reprinted from reference 10, with permission.

nese encephalitis and those who recently had dengue or St. Louis encephalitis because of the close antigenic relationships between the flaviviruses[16]; therefore, it is critical to obtain a travel and immunization history for accurate interpretation of serologic results. The plaque reduction neutralization test may help distinguish these false-positive results.[1] Clinicians should also note that virus isolation from serum or CSF is often unsuccessful because of low viremia.[23] Viremia may last longer in patients taking immunosuppressive drugs or with advanced malignancies.

Another consideration is that patients in endemic areas may be asymptomatic but still have IgM antibodies for at least six months. If these patients become ill from an unrelated infection and are tested for WNV antibodies, the diagnosis may be confusing.[16] In these patients, acute infection may be confirmed by an increase in WNV-specific neutralizing antibody titer in the serum.

Clinically, patients may have mild leukocytosis or mild leukopenia.[16] Mild hyponatremia can occur and is more common in patients with encephalitis. CSF findings include mild pleocytosis with lymphocytic predominance, mild to moderate protein elevation, and normal glucose levels.[16,23] Computed tomography usually shows no sign of acute disease.[16]

Treatment

Currently, there is no established treatment for WNV infection. Treatment consists only of supportive and symptomatic care, including respiratory support, intravenous fluids, and prevention of secondary infections. Human vaccines for WNV are being developed.[16,30,40] The vaccines use the protype flavivirus, yellow fever 17D, as a live vector and replace the genes encoding the premembrane and envelope protein of yellow fever 17D vaccine with the corresponding genes of WNV. The resulting virus contains the antigens responsible for protection against WNV.[40] High-dose ribavirin and interferon alfa-2b have shown efficacy in vitro.[41] Interferon alfa (a glycoprotein cytokine) has produced positive results in open trials in patients with Japanese encephalitis and St. Louis encephalitis.[41,42] However, a randomized, double-blind, placebo-controlled trial of patients with Japanese encephalitis treated with interferon alfa (10 million units/m^2 for seven days) demonstrated no benefits.[43] High-dose ribavirin did not show any benefit during the WNV outbreak in Israel in 2000.[14] The use of corticosteroids, antiseizure medications, and osmotic agents has yet to be studied in the management of West Nile encephalitis. An intravenous immunoglobulin preparation containing a high titer of anti-WNV antibodies has been successfully used in an immunosuppressed patient[44] and demonstrated prophylactic and therapeutic efficacy in mice.[45] Larger studies are required to establish the

therapeutic role of immunoglobulins in patients with WNV infection.[23]

Prevention

Prevention can be divided into three general categories: (1) preventing mosquitoes infected with WNV from biting humans, (2) reducing the number of mosquitoes, and (3) public education.[46,47] Of these, mosquito control is the most effective means to prevent WNV transmission.

Insect repellent should be applied to exposed skin and clothing before going outdoors. The most effective repellents contain DEET (N,N-diethyl-m-toluamide). Products with a DEET concentration of >50% do not increase the length of protection, and products containing no more than 10% DEET are appropriate for children 2–12 years of age. Repellents should not be placed on the hands of children, and care should be taken to avoid the products' contact with their mouth and eyes.[47] Protective clothing, such as long sleeves, long pants, and socks, should be worn whenever possible when outdoors. Clothing should be sprayed with repellents since mosquitoes can bite through thin clothing. Stay indoors between dusk and dawn, as that is the peak time for mosquito bites.

Environmental measures include creating barriers, such as properly installed window and door screens, and minimizing mosquito-breeding areas, by draining standing water. Water should be routinely emptied from flowerpots, clogged rain gutters, swimming pool covers, pet bowls, discarded tires, buckets, barrels, cans, and other items that collect water. Local health authorities should be alerted to potential mosquito-breeding sites in areas, such as storm sewers, ditches, and abandoned properties with standing water. Dead or dying birds and horses that are suspected of being infected with WNV should also be reported. Each state and county reports this information to the Centers for Disease Control and Prevention (CDC).[47]

Public education about WNV and the potential health and environmental impact of mosquito control is essential. Surveillance of bird deaths, mosquito-breeding sites, and human cases of WNV is also critical for improving infection-control strategies.[46,47] Additional information about WNV is available from CDC's Division of Vector-Borne Infectious Diseases at www.cdc.gov/ncidod/dvbid/westnile/city_states.htm.

Since this manuscript was accepted for publication, more recent data on the spread of WNV have been released. In 2003, 9,306 human infections and 210 deaths were associated with WNV.[48] In addition, 11,597 dead birds tested positive for WNV infection. CDC has recently recommended that pregnant women with meningitis, acute flaccid paralysis, or unexplained fever in an area of WNV transmission should have serum (and CSF, if clinically indicated) tested for the presence of WNV IgM antibody.[49] This new recommendation also suggests that pregnant women living in areas with WNV-infected mosquitoes apply insect repellent to clothes and skin, wear clothing to protect against mosquito bites, and avoid the outdoors during peak mosquito-feeding times. Concern for WNV infection also applies to infants born to mothers who were diagnosed or suspected to have been infected with WNV during pregnancy. Clinical and laboratory evaluations should be conducted as indicated.[49]

Conclusion

There is no established treatment for WNV infection. Currently, prevention and control are the only measures that help decrease the morbidity and mortality associated with WNV infection. As the number of cases escalates and the geographic distribution of WNV infection widens, the epidemic will continue to pose a major challenge to clinicians in the coming years. There is an urgent need for more research on the pathogenesis and treatment of WNV infection.

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