## **West Nile Virus**

# History

The West Nile Virus (WNV) was first isolated from a woman in the West Nile District of Uganda in 1937. The virus, which can cause an encephalitis (inflammation of the brain) in animals and in some cases, humans, has been found in Africa, western Asia, the Middle East and the Mediterranean region of Europe.

The virus was first isolated in the United States from a dead crow on September 14, 1999, by the National Veterinary Services Laboratory (NVSL). The crow was found by a zoo veterinarian in New York City. The virus was later identified as WNV and was confirmed as the cause of a human encephalitis case that occurred in August, also in New York City. The virus was also found in Connecticut, New Jersey and one county in Maryland. In 1999 the virus was found in wild birds, mosquitos, horses and people. There have been 16 human deaths caused by the WNV since 1999.

## The Virus

West Nile Virus is classified as an arbovirus. Arboviruses are viruses that are transmitted by blood feeding insects. Interestingly, the word arbovirus comes from the combination of the words arthropod borne virus. It is specifically a mosquito-borne virus affecting both the central and peripheral nervous system of people and animals The virus belongs to the flavivirus family (flaviviradae) which includes the viruses that cause Japanese encephalitis, St. Louis encephalitis, Bovine Virus Diarrhea, Pestivirus diseases such as Hog Cholera and Border Disease, Hepatitis C, Dengue Fever and Yellow Fever.

# Surveillance for WNV

Testing of dead wild birds is probably the best surveillance method for WNV coupled with sentinel chicken flocks. In 2001, 15 dead crows, 14 magpies, 2 ravens and 1 unidentified bird tested negative for WNV in Colorado. Sentinel Chicken Flocks were maintained in the Colorado counties of Adams, Boulder, Delta, Jefferson, Larimer, Logan, Mesa, Moffat, Otero, Prowers, Pueblo, Weld and Yuma in 2001. The virus has not been identified in Colorado as of May 26, 2002. For more information visit the Colorado Department of Public Health site at <a href="http://www.cdphe.state.co.us/dc///////zoonosis/

Dead birds should be reported to your local health department. Horses exhibiting neurological signs that are suspected of having an encephalomyelitis must be reported to the Colorado State Veterinarian's Office (303-239-4161)

# The Disease

West Nile Virus (WNV) is a mosquito-borne virus, characterized by inflamation of the brain and spinal cord

Most WNV infections in people do not cause any illness. Those with mild WNV encephalitis can run a fever and have head and body aches. They can also develop skin rashes, experience extreme tiredness and have swollen lymph nodes. People with more serious infections can develop a stiff necks, go into a stupor, become disoriented, develop tremors, run a much higher temperature, become paralyzed, convulse, go into a coma and in some cases die. The incubation period or the time between exposure to the infected mosquito and the onset of symptoms, ranges from 5 to 15 days.

## Horses

The most current USDA Animal and Plant Health Inspection Service statistics show that 651 verified clinical cases of WNV infections were reported in horse in 130 counties and 20 states during the year 2001. For those cases where an outcome was reported, 33.2% of the horses died or were euthanized. Florida, Georgia and New Jersey had the greatest number of deaths (link to USDA statistics)(http://www.aphis.usda.gov/oa/wnv/wnvstats.html)

Similar to people, many horses will not become clinically ill when infected with the WNV (approximately 1 out of 20). Others may become depressed and listless. Although affected horses do not usually develop an elevated temperature, they can develop neurological symptoms of head tilt, muscle tremors, incoordination and partial paralysis. They may appear to be experiencing weakness of the limbs with knuckling over and a stumbling gait. They may go down and be unable to rise. Clinical signs similar to rabies in horses have been reported by some.

A review of clinical WNV cases in the US during 1999 and 2001 have not shown any breed predilection. The occurrence rate has been slighter higher in males (53.7%) than females. It also appears that all ages are infected, but more older horses become ill than younger ones, and of the horses that become ill, those that die are older than those that recover. Eighty four percent of the cases have occurred in the south. Of the horses that became ill, 68 percent of horses between the ages of 25 and 29 died compared to a mortality rate of 24 percent of those 4 and under.

Equine WNV cases in the US have occurred between July and December, however, a majority of these cases occurred between late August and early November.

The case-control study also showed that infected horses were less likely to be housed in stalls at night and more likely to be used for pleasure purposes. It was also observed that the affected horses where more likely to be located within ½ mile of a blackbird roost or sites of waterfowl congregation. It was also observed that cases were clustered in areas.

# Poultry and Other Animals

While the West Nile Virus primarily affects horses, it has also been observed in bats, squirrels, chipmunks, raccoons, skunks, rabbits and cats. Although there is a published report of WNV isolated from a dog in southern Africa (Botswana) in 1982, no cases have been documented in dogs in the US.

WNV infections have been documented in sheep, cattle and pigs in Africa and Eurasia. Most infections were without clinical signs and animals developed antibodies to the virus. However, in laboratory studies sheep infected with WNV exhibited fever, abortion and encephalitis.

Chickens can be infected with WNV and not become sick. However, natural disease due to the virus has been reported in domestic geese, ducks, pigeons and chickens. Israel has destroyed thousands of geese infected with WNV and began a vaccination program to prevent the spread of the disease.

## Birds

Many species of birds usually do not show any clinical signs when infected with WNV. In the United States since WNV was first detected, it has been found in over 80 species of birds. Of these, 60 species were free ranging at the time of the positive test. WNV does appear to be particularly lethal for the American crow, fish crow and blue jays. Other Corvids, like magpies and ravens as well as other species of crows and jays, may be equally susceptible. There is also concern that the virus poses a very serious risk for threatened species of birds like the whooping crane, scrub jay and wood stork. For a more complete list of wild birds, you can visit the USGS National Wildlife Health Center's Web page at <a href="http://www.nwhc.usgs.gov/whats\_new/wha/wha0102.html">http://www.nwhc.usgs.gov/whats\_new/wha/wha0102.html</a>

#### Treatment

There are no specific treatments for the virus. Infected animals and people are given good supportive care while the disease runs it's course. Infected animals should be treated by your veterinarian..

# Spread of the Disease

There are several schools of thought about the spread of the disease. Some epidemiologists have implicated migratory birds in the spread of the virus. While the virus is deadly to Corvids, most species of migratory birds are not seriously affected by the virus. During the winter months, while in Mexico and Central America, migrating birds pick up the virus and carry it back to new areas in the United States. However, if maps of the occurrence of the disease are viewed (**link to maps** 2000 & 2001), there appears to be a steady movement of the disease across the US from east to west. This appears to support the second group of epidemiologists who feel that the disease is being spread by infected mosquitos that overwinter. Some epidemiologists are predicting that the virus will reach Texas, the Rocky Mountains and possibly the West Coast this year (2002). Infected birds serve as the reservoir of infection. Humans and horses can become infected with the WNV. However, the infections are usually considered to be dead end infections since there is little virus circulating in the blood stream, posing little risk for infection to other people and horses.

# Mosquitos and Transmission

Wild birds are the primary vertebrate host for WNV and act as an amplifying host. Because of the high viremias produced, they serve as a reservoir for the virus. Mosquitos that feed on the infected wild birds pickup and carry the virus in their salivary glands. The virus carrying mosquito then transmits the disease to susceptible humans, animals and other birds when they feed again. The virus has been found in 22 species of mosquitos including - *Culex sp.*, *Culiseta melanura*, *Aedes triseriatus*, *Aedes cantator*, *Aedes japonicus*, *Aedes vexans*, *Aedes albopictus*, *Aedes trivitfatus*, *Anopholese punctpennis*, and *Psorophora ferox*. In the United States, *Culex sp.* have been primarily responsible for the spread of the disease. It is not known what role other blood sucking insects and ticks might play in the transmission of the disease.

# Pathogenesis

After the virus is introduced into the host from an infected mosquito bite, it multiplies in the tissues and lymph nodes near the entry site. The virus gains access to the blood stream through the lymphatics. A viremia (viruses in the bloodstream) occurs relatively early in the course of the disease. Studies suggest that infection of the brain occurs when the virus crosses the blood brain barrier, which is different from the St. Louis Encephalitis Virus where neuroinvasion occurs through the olfactory nerve..

## Prevention and Control

There are several strategies that can be employed to lower the risk of infection. For people, the strategy is limited to mosquito control and minimizing their exposure to mosquitos. In addition, Fort Dodge Animal Health has received a conditional license to manufacture a killed WNV vaccine to be used "as a aid in the prevention of diseases caused by the WNV" in horses.

# Mosquito Control

Effective mosquito control has to be a community wide effort. Individual roles in this effort are aimed at eliminating standing water that provide excellent breeding sites for the mosquitos. Dispose of, cover or turn upside down unused items like buckets, wheelbarrows, plastic wading pools, cans, pots, old tires, and other containers capable of holding water. Drill holes in feed pans, trash containers, tire swings, holes in trees, and all other outdoor containers to prevent the accumulation of water. Fill or drain ground depressions that hold water for more than four days. Stock tanks, ornamental pools and ponds should be aerated or stocked with mosquito fish (*Gambusia affinis*). Excavation of shallow ponds to make them deeper can also assist in eliminating mosquito breeding sites.

Individuals can also reduce their risks by limiting their exposure by spending less time outside in the early evening. Long sleeved shirts and full length pants should be worn when outside and mosquito repellents should be applied to areas of bare skin.

Community wide efforts will have a much greater effect on controlling mosquitos. In addition to eliminating breeding sites, communities can initiate mosquito control using mosquito larvicides to treat breeding habitats. *Bacillus thuringiensis* (*Bt*) *israelensis* and *Bacillus sphaericus* are

considered to be environmentally friendly, naturally occurring, soil borne bacteria used to control mosquito larvae (caterpillar-like). Since they must be ingested to work, they are effective only in the larval stages, since this is the stage when most of the feeding occurs. When the bacteria are ingested, they grow in numbers and produce toxins that paralyze the gut causing the larvae to starve to death.

Oils and monomolecular films are also used in mosquito larva control by creating barriers on the top of water that block larval access to oxygen effectively drowning them.

Methoprene is a chemical larvicide that mimics an Insect Growth Regulating Hormone (IGRH). These compounds have a very low level of toxicity for mammals, fish and bees.

Temephos is an organophosphate pesticide registered for control of mosquito larvae. It is considered if used according to labeled directions. It is an important tool to manage larval resistance to the bacterial larvicides.

If efforts to eliminate mosquito habitat and mosquito larvicides fail to control the mosquito population, efforts may have to be directed at killing adult mosquitos. Malathion and Naled are organophosphates registered and labeled for use in killing adult mosquitos. They can be used in agricultural applications, residential gardens and public areas. They can be safely used if applied at labeled rates following the labeled directions.

Pyrethroids a synthetic insecticides. Their mechanisms of action are similar to naturally occurring insecticides produced by Chrysanthemum flowers named pyrethrins. Permethrin, resmethrin and sumithrin are pyrethroids used to control adult mosquitos. These compounds are classified as Restricted Use Pesticides and can only be used by certified applicators. Pyrethroids have a low toxicity for mammals and are practically non toxic to birds. However, because they are toxic to fish and bees, the can only be applied by certified applicators.

## Vaccination

In addition to the mosquito-related prevention measures outlined above, there is one additional action that can be taken to help prevent illness in horses caused by WNV infection. On August 1, 2001, a conditional license was issued by the USDA-APHIS' Center for Veterinary Biologics for an equine WNV vaccine to Fort Dodge Animal Health in Ames, Iowa. The vaccine is a killed virus product that initially has a one-year license. Conditional licensing means that the product has been shown to be safe, pure, and have a reasonable expectation of efficacy in preventing illness caused by WNV. Because use of this vaccine is restricted to veterinarians, you need to contact your veterinarian to find out more about its availability and use in your area. The vaccine has been approved for use in Colorado. The vaccine label recommends giving two intramuscular doses of 1 milliliter each, 3 to 6 weeks apart initially with revaccination annually.

The labeled indications for the vaccine are for use in healthy horses "as an aid in prevention of disease caused by West Nile Virus".

Over 1 million doses have been distributed to date. There have been no reported adverse events regarding the WNV vaccine to the USDA Center for Veterinary Biologics at the time this article was written. However, it appears that vaccination failure can occur. A review of WNV cases in vaccinated horses reported 5 cases in horses that occurred at least 20 days after the second dose of vaccine (20,21,27,33,37 days)(R. L. Crom, DVM, USDA, APHIS, VS)

Eastern Equine Encephalomyelitis (EEE), Western Equine Encephalomyelitis (WEE), and Venezuelan Equine Encephalomyelitis (VEE) are caused by alphaviruses. West Nile is a flavivirus so vaccinations for EEE, WEE, and VEE provide no cross-protection for the WNV.

# Diagnosis and Testing

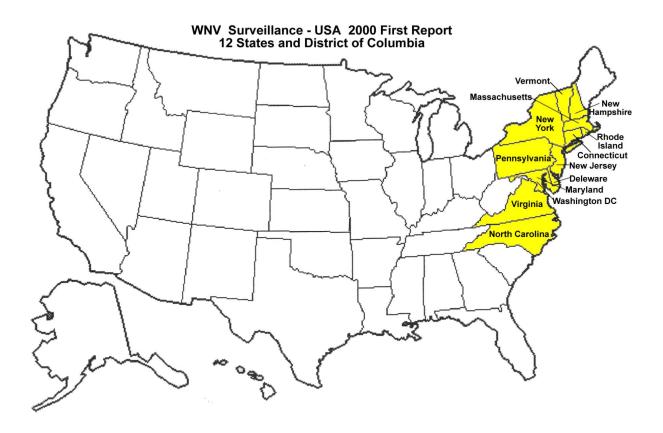
Through collaboration with the Centers for Disease Control and Prevention (CDC) and the National Veterinary Services Laboratory (NVSL), Colorado Department of Agriculture's (CDA) Rocky Mountain Regional Animal Health Laboratory (RMRAHL) is conducting an Immunoglobulin M (IgM) antibody capture enzyme-linked immunosorbent assay (MAC-ELISA) for West Nile Virus (WNV). IgM antibodies are the first antibodies produced in response to primary exposure or infection to the WNV. In experimental studies conducted at NVSL's Diagnostic Virology Laboratory (DVL), infected horses usually start producing detectable IgM antibody 7 to 10 days after infection. In addition, their work with field cases indicates that most horses are positive for IgM antibodies by the start of clinical symptoms.

Membrane receptor preparation (MRP) binding assays have shown that vaccinated horses do not develop an IgM antibody response after either the 1<sup>st</sup> or 2<sup>nd</sup> dose. Only IgG antibody titers have been detected in vaccinated horses. Positive test titers from the RMRAHL lab tests\*\* should be indicative of exposure to the West Nile Virus and not from vaccination.

<u>Questions and answers</u> Click the link to go to the CDC Q&A site (http://www.cdc.gov/ncidad/dvbid/westnile/q&a.htm)

\*\* RMRAHL is conducting the MAC-ELISA Monday through Wednesday on equine serum. Test results are available in 48 hours. The fee for this service is \$4.75 per sample. Samples may be sent to the:

CDA - RMRAHL 2331 West 31<sup>st</sup> Ave. Denver, CO 80211 For more information regarding WNV testing or other services offered by RMRAHL, please phone  $(303)\,477\text{-}0049$ .



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