West Nile Fever—a Reemerging Mosquito-Borne Viral Disease in Europe

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West Nile virus causes sporadic cases and outbreaks of human and equine disease in Europe (western Mediterranean and southern Russia in 1962-64, Belarus and Ukraine in the 1970s and 1980s, Romania in 1996-97, Czechland in 1997, and Italy in 1998). Environmental factors, including human activities, that enhance population densities of vector mosquitoes (heavy rains followed by floods, irrigation, higher than usual temperature, or formation of ecologic niches that enable mass breeding of mosquitoes) could increase the incidence of West Nile fever.

The 1996-97 outbreak of West Nile fever in and near Bucharest, Romania, with more than 500 clinical cases and a case-fatality rate approaching 10% (1-3), was the largest outbreak of arboviral illness in Europe since the Ockelbo-Pogosta-Karelian fever epidemic caused by Sindbis virus in northern Europe in the 1980s. This latest outbreak reaffirmed that mosquito-borne viral diseases may occur on a mass scale, even in temperate climates.

West Nile virus is a member of the Japanese encephalitis antigenic complex of the genus *Flavivirus*, family *Flaviviridae* (4). All known members of this complex (Alfuy, Japanese encephalitis, Kokobera, Koutango, Kunjin, Murray Valley encephalitis, St. Louis encephalitis, Stratford, Usutu, and West Nile viruses) are transmissible by mosquitoes and many of them can cause febrile, sometimes fatal, illnesses in humans.

West Nile virus was first isolated from the blood of a febrile woman in the West Nile district of Uganda in 1937 (5) and was subsequently isolated from patients, birds, and mosquitoes in Egypt in the early 1950s (6-7). The virus was soon recognized as the most widespread of the flaviviruses, with geographic distribution including Africa and Eurasia. Outside Europe (Figure), the virus has been reported from Algeria, Asian Russia, Azerbaijan, Botswana, Central African Republic, Côte d'Ivoire, Cyprus, Democratic Republic of Congo (former Zaire),

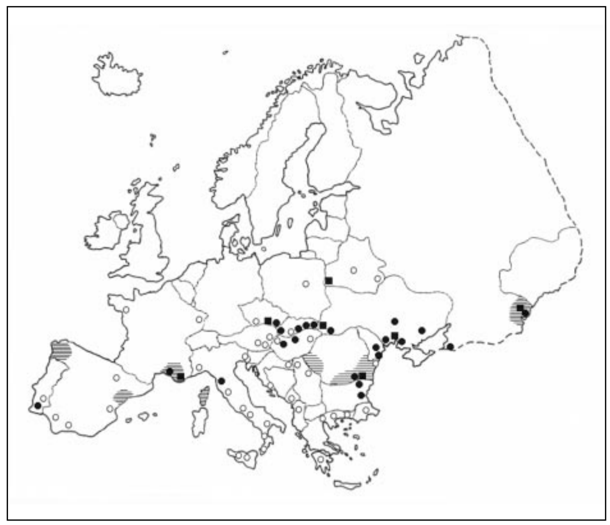

Figure. European distribution of West Nile virus, based on the virus isolation from mosquitoes or vertebrates, including humans (black dots), laboratory-confirmed human or equine cases of West Nile fever (black squares), and presence of antibodies in vertebrates (circles and hatched areas).

Egypt, Ethiopia, India, Israel, Kazakhstan, Madagascar, Morocco, Mozambique, Nigeria, Pakistan, Senegal, South Africa, Tajikistan, Turkmenia, Uganda, and Uzbekistan. Furthermore, West Nile virus antibodies have been detected in human sera from Armenia, Borneo, China, Georgia, Iraq, Kenya, Lebanon, Malaysia, the Philippines, Sri Lanka, Sudan, Syria, Thailand, Tunisia, and Turkey (8-10). Kunjin virus is closely related to West Nile virus (11,12), representing a counterpart or subtype for Australia and Southeast Asia; some West Nile

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virus seroreactions in Southeast Asia may, in fact, represent antibodies to Kunjin virus.

West Nile Virus Ecology

Arthropod Vectors

Mosquitoes, largely bird-feeding species, are the principal vectors of West Nile virus. The virus has been isolated from 43 mosquito species, predominantly of the genus *Culex* (Table 1). In Africa and the Middle East, the main vector is *Cx. univittatus* (although *Cx. poicilipes, Cx. neavei, Cx. decens, Aedes albocephalus,* or *Mimomyia* spp. play an important role in certain areas). In Europe, the principal vectors are *Cx. pipiens*, *Cx. modestus,* and *Coquillettidia richiardii*, and in Asia, *Cx. quinquefasciatus*, *Cx. tritaeniorhynchus,* and *Cx. vishnui* predominate. Successful experimental transmission of the virus has been described in *Culiseta longiareolata*, *Cx. bitaeniorhynchus,* and *Ae. albopictus* (8,13). Transovarial transmission of the virus has been demonstrated in *Cx. tritaeniorhynchus*, *Ae. aegypti*, and *Ae. albopictus,* though at low rates.

Virus isolations have occasionally been reported from other hematophagous arthropods (e.g., bird-feeding argasid [soft] or amblyommine [hard] ticks) (Table 1), and experimental transmission has been observed in *Ornithodoros savignyi*, *O. moubata*, *O.maritimus*, *O. erraticus*, *Rhipicephalus sanguineus*, *R. rossicus*, *Dermacentor reticulatus,* and *Haemaphysalis leachii* (8,13).

Vertebrate Hosts

Wild birds are the principal hosts of West Nile virus. The virus has been isolated from a number of wetland and terrestrial avian species in diverse areas (7-10,14-16). High, long-term viremia, sufficient to infect vector mosquitoes, has been observed in infected birds (7,17,18). The virus persists in the organs of inoculated ducks and pigeons for 20 to 100 days (18). Migratory birds are therefore instrumental in the introduction of the virus to temperate areas of Eurasia during spring migrations (12,14-16,19).

Rarely, West Nile virus has been isolated from mammals (*Arvicanthis niloticus*, *Apodemus flavicollis*, *Clethrionomys glareolus*, sentinel mice and hamsters, *Lepus europaeus*, *Rousettus leschenaulti*, camels, cattle, horses, dogs, *Galago senegalensis*, humans) in enzootic foci (8-10). Mammals are less important than birds in maintaining transmission cycles of the virus in ecosystems. Only horses and lemurs (20) have moderate viremia and seem to support West Nile virus circulation locally. Frogs (*Rana ridibunda*) also can harbor the virus, and their donor ability for *Cx. pipiens* has been confirmed (21).

Transmission Cycles

Although Palearctic natural foci of West Nile virus infections are mainly situated in wetland ecosystems (river deltas or flood plains) and are characterized by the bird-mosquito cycle, argasid and amblyommine ticks may serve as substitute vectors and form a bird-tick cycle in certain dry and warm habitats lacking mosquitoes. Even a frog-mosquito cycle (21) may function under certain circumstances.

In Europe, West Nile virus circulation is confined to two basic types of cycles and ecosystems: rural (sylvatic) cycle (wild, usually wetland birds and ornithophilic mosquitoes) and urban cycle (synanthropic or domestic birds and mosquitoes feeding on both birds and humans, mainly *Cx. pipiens/molestus)*. The principal cycle is rural, but the urban cycle predominated in Bucharest during the 1996-97 outbreak (2,3). Circulation of West Nile fever in Europe is similar to that of St. Louis encephalitis in North America, where the rural cycle of exoanthropic birds—*Cx. tarsalis* alternates with the urban cycle of synanthropic birds—*Cx. pipiens/ quinquefasciatus*.

West Nile Fever in Humans and Other Vertebrates

Humans

West Nile fever in humans usually is a febrile, influenzalike illness, characterized by an abrupt onset (incubation period is 3 to 6 days) of moderate to high fever (3 to 5 days, infrequently biphasic, sometimes with chills), headache (often frontal), sore throat, backache, myalgia, arthralgia, fatigue, conjunctivitis, retrobulbar pain, maculopapular or roseolar rash (in approximately half the cases, spreading from the trunk to the extremities and head), lymphadenopathy, anorexia, nausea, abdominal pain, diarrhea, and respiratory symptoms (9). Occasionally (<15% of cases), acute aseptic meningitis or encephalitis (associated with neck stiffness, vomiting, confusion, disturbed consciousness, somnolence, tremor of extremities, abnormal reflexes,

Table 1. Isolations of West Nile virus from hematophagous arthropods (7-10)

Species	No.	Countries
Mosquitoes		
Culex antennatus[a]	6	Egypt, Madagascar
decens group	8	Madagascar
ethiopicus	1	Ethiopia
guiarti	1	Côte d"Ivoire
modestus	3	France, Russia
neavei	4	Senegal, South Africa
nigripes	1	Central African Republic
perexiguus	1	Israel
perfuscus group	3	Central African Republic, Senegal
pipiens[a]	7	South Africa, Egypt, Israel, Romania, Czechland, Bulgaria[b]
poicilipes	29	Senegal
pruina	1	Central African Republic
quinquefasciatus[a]	7	India, Pakistan, Madagascar
scottii	1	Madagascar
theileri[a]	4	South Africa
tritaeniorhynchus[a]	3	Pakistan, India, Madagascar
univittatus[a]	51	Egypt, Israel, South Africa, Madagascar
vishnui[a] group	6	India, Pakistan
weschei	1	Central African Republic
sp.	3	Egypt, Algeria, Central African Republic
Coquillettidia metallica	1	Uganda
microannulata	1	South Africa
richiardii	5	South Russia, Bulgaria[b]
Mansonia uniformis	1	Ethiopia
Aedes aegypti[a]	1	Madagascar
africanus	1	Central African Republic
albocephalus	35	Madagascar
albothorax	1	Kenya
cantans	7	Slovakia, Ukraine, Bulgaria[b]
caspius[a]	1	Ukraine
circumluteolus	2	South Africa, Madagascar
excrucians	1	Ukraine
juppi+caballus	1	South Africa
madagascarensis	1	Madagascar
vexans	3	Senegal, Russia
Anopheles brunnipes	1	Madagascar
coustani	1	Israel
maculipalpis	1	Madagascar
maculipennis	3	Portugal, Ukraine
subpictus	1	India
sp.	1	Madagascar
Mimomyia hispida	8	Senegal
lacustris	4	Senegal
splendens	6	Senegal
sp.	2	Senegal
Aedeomyia africana	1	Senegal
Soft ticks		
Argas hermanni[a]	3	Egypt
Ornithodoros capensis[a]	5	Azerbaijan
Hard ticks		
Hyalomma marginatum	5	Astrakhan, Azerbaijan
detritum	1	Turkmenistan
Rhipicephalus turanicus	1	Azerbaijan
muhsamae	1	Central African Republic
Amblyomma variegatum	1	Central African Republic
Dermacentor marginatus[a]	1	Moldavia

[a]Experimental transmission of the virus also demonstrated.
[b]Detected in mosquitoes by immunofluorescence assay.

convulsions, pareses, and coma), anterior myelitis, hepatosplenomegaly, hepatitis, pancreatitis, and myocarditis occur. Laboratory findings involve a slightly increased sedimentation rate and a mild leukocytosis; cerebrospinal fluid in patients with central nervous system involvement is clear, with moderate pleocytosis and elevated protein. The virus can be recovered from the blood for up to 10 days in immunocompetent febrile patients, as late as 22 to 28 days after infection in immunocompromised patients; peak viremia occurs 4 to 8 days postinfection. Recovery is complete (less rapid in adults than in children, often accompanied by long-term myalgias and weakness), and permanent sequelae have not been reported. Most fatal cases have been recorded in patients older than 50 years. Many of the West Nile fever symptoms have been reproduced in volunteers with underlying neoplastic disease who had been inoculated with virus to achieve pyrexia and oncolysis (22).

Hundreds of West Nile fever cases have been described in Israel and South Africa. The largest African epidemic, with approximately 3,000 clinical cases, occurred in an arid region of the Cape Province after heavy rains in 1974 (23). An outbreak with approximately 50 patients, eight of whom died, was described in Algeria in 1994 (1). Other cases or outbreaks have been observed in Azerbaijan, Central African Republic, Democratic Republic of Congo (former Zaire), Egypt, Ethiopia, India, Madagascar, Nigeria, Pakistan, Senegal, Sudan, and in a few European countries.

Horses

Equine disease, called Near Eastern equine encephalitis in Egypt and lourdige in France, was observed and experimentally reproduced as fever and diffuse encephalomyelitis with a moderate to high fatality rate in Egypt (24), France (c. 50 cases in 1962-65) (25), Italy (14 cases in 1998, six died or were euthanised) (R. Lelli, G. Ferrari, pers. comm.), Portugal (26) and Morocco (42 of 94 affected horses died) (27). In the 1960s, the biphasic, encephalomyelitic form, which causes staggering gait and weakness to paralysis of the hind legs, was apparent among infected semiferal horses in Camargue (25).

Other Mammals

Inoculation of sheep with West Nile virus results in fever, abortion in pregnant ewes, and rare encephalitis, in contrast to the asymptomatic infection seen in pigs and dogs (9,28). Rabbits, adult albino rats, and guinea pigs are resistant to West Nile virus infection, but laboratory mice and Syrian hamsters are markedly susceptible; they often become ill with fatal encephalitis, even when inoculated peripherally (8). Adult rodents stressed or immunosuppressed by cold, isolation, cyclophosphamide, corticosterone, or bacterial endotoxin contract fatal encephalitis, even when an attenuated viral strain is given (29). Inoculation of rhesus and bonnet monkeys (but not cynomolgus monkeys or chimpanzees) causes fever, ataxia, and prostration with occasional encephalitis, tremor of extremities, pareses, or paralysis. Infection may be fatal or cause long-term virus persistence in survivors (5,6,30).

Birds

Birds usually do not show any symptoms when infected with West Nile virus. However, natural disease due to the virus has been observed in a pigeon in Egypt (7), and inoculation of certain avian species (e.g., pigeons, chickens, ducks, gulls, and corvids) causes occasional encephalitis and death or long-term virus persistence (7,10,17,18). Chick embryos may be killed by the virus (8).

West Nile Virus and Fever in Europe

In Europe, the presence of West Nile virus was indicated in 1958, when two Albanians had specific West Nile virus antibodies (31). The first European isolations of the virus were recorded in 1963 from patients and mosquitoes in the Rhône Delta (32) and from patients and *Hyalomma marginatum* ticks in the Volga Delta (33,34). West Nile virus was subsequently isolated in Portugal (35), Slovakia (36), Moldavia (37), Ukraine (38), Hungary (39), Romania (2), Czechland (40), and Italy (V. Deubel, G. Ferrari, pers. comm.).

The incidence of West Nile fever in Europe is largely unknown. In the 1960s, cases were observed in southern France (25), southern Russia (41), Spain (26), southwestern Romania (42), in the 1970s, 1980s, and 1990s in Belarus (43), western Ukraine (44), southeastern Romania (1,2), and Czechland (45). West Nile fever in Europe occurs during the period of maximum annual activity of mosquito vectors (July to September) (Table 2).

Table 2. West Nile Virus in Europe, 1960-1998

Country	Year	Species infected	HI[a] (%)	Neutralization (%)	Ref.
Portugal, southern	1967-1970	Cattle, sheep	15		26, 35
		Horses	29		
		Humans	3		
		Mosquitoes (1 isolate)			
		Wild birds	5		
Spain, northern	1979	Rodents	3		
Northwestern	1960s	Humans	17	17	26
Doñana National Park		Humans	+		
Ebro Delta	1979	Epidemic of influenza-like illness	8-30		
France, southern	1962	Humans, 10 severe cases (2 isolates)	19		25,32,46
		Horses, 50 encephalomyelitis cases (1 isolate)	9	30	
Camargue	1962-1965	Mosquitoes (2 isolates)			
		Wild birds	6		
	1975-1980	Humans	5		
		Horses	2		
Corsica	1965	Humans	30	55	
Italy, Tuscany	1998	Horses, 14 cases, 2 fatal (1 isolate)		40	47
Northeastern	1967-1969	Migrating birds	10		
		Humans	5		[b]
Northwestern		Humans	23		
Central		Humans	2-8		
		Domestic mammals	8		
		Rodents	8		
		Chickens	20		
Southern		Humans	2-5		
		Goats	2-13		
	1981	Rodents	1		
Former Yugoslavia					
Serbia		Humans	1-8		48
Croatia		Humans	1-3		
Montenegro		Humans	1		
Bosnia, Kosovo		Humans	1		
Albania	1958	Humans		2	31,49
		Domestic animals			
Greece	1970-1978	Humans	1-27	1	50,51
		Domestic animals	7		
		Rabbits	4		
		Birds	22		
Bulgaria	1960-1970	Humans	3		52,53
Eastern		Wetland birds	2	10	
		Domestic animals	1		
	1978	Mosquitoes (virus detected)			
Romania					1-3,42,54,55
Bucharest, SE lowlands	1996	Humans, 453 clin. cases, 9% fatality rate (1 isolate)	17		
	1997	Human, 14 cases, 2 fatal			
		Domestic & wild mammals	2-23		[c]
		Dogs	19-45		
		Wild birds	22		
	1966-70	Mosquitoes (1 isolate)			
Banat (SW)		Humans (cases)	17		
Southern	1980-1995	Humans	2-12		
Hungary	1970s	Rodents (2 isolates)			39,56
		Cattle	4-9		
		Humans	4-6		
Slovakia	1972	Mosquitoes (1 isolate)			16, 36, 57-60
	1970-1973	Migrating birds (4 isolates)		1-13	
		Game animals		1-8	
		Cattle, dogs	8		
		Sheep	1		
		Pigeons	5		
		Humans	1-4		
Austria	1964-1977	Wetland passerines	1-3		61,62
		Reptiles			
		Wild mammals			
		Domestic animals	7-33		
		Humans	1-6		
Czechland	1978	Domestic animals	2		40,45,63-67
Southern Bohemia	1978	Hares	5		
Southern Moravia	1980s	Game animals	8		
	1985	Wetland birds	4-10		
	1990	Cormorants	10		
	1997	Mosquitoes (1 isolate)			
		Humans (5 cases)		2	
Poland, near Warsaw	1996	Sparrows	3-12		68
Belarus	1977	Humans (cases in Brest area)	1		43
	1972-1973	Wild birds	3		
Ukraine					38,44,69
Southern, western		Birds (7 isolates), mosquitoes (3 isolates)			
Southern	1970s	Human cases (4 isolates)			
Western	1985	Humans, 38 cases, encephalitis in 16			
Moldavia	1970s	Ticks, mosquitoes (several isolates)			37, 70
		Humans	3		
Russia,					33,34,41,55
Volga Delta	1963-1968	Humans (>10 cases, 3 isolates)	7-31		
		Ticks (4 isolates)			
		Water birds (2 isolates)	4-59	2-11	
		Mosquitoes (2 isolates)			

[a]Hemagglutination inhibition.
[b]Q. Ferrari, R. Lelli, pers. comm.
[c] C. Ceianu, pers. comm.

The Future

West Nile virus can cause sporadic human cases, clusters, or outbreaks of West Nile fever, even in temperate Europe. Environmental factors, including human activities that enhance vector population densities (irrigation, heavy rains followed by floods, higher than usual temperatures, and formation of ecologic niches enabling the mass breeding of mosquitoes) allow the reemergence of this mosquito-borne disease. For instance, global warming scenarios hypothesize warmer, more humid weather that may produce an increase in the distribution and abundance of mosquito vectors (71). Surveillance for West Nile fever (monitoring population densities and infection rates of principal vectors, serosurveys on vertebrates and exposed human groups, and routine diagnosis of human infections) should therefore be carried out in affected areas.

The mechanism of West Nile virus persistence in disease-endemic foci of temperate Europe presents a challenge for further research. General hypotheses of how an arbovirus could overwinter under adverse climatic conditions have already been postulated (72). The virus could persist in hibernating female *Culex* spp.; transovarially infected *Culex* spp. progeny; or chronically infected vertebrate hosts, perhaps birds or frogs. Alternatively, the virus may be reintroduced by chronically infected migratory birds from tropical or subtropical foci at irregular intervals. These issues have to be addressed, because present data substantiate all particular mechanisms and their combinations. For instance, the hibernating vector idea has been supported by a few field and experimental data on female *Cx. univittatus* (7,73). On the other hand, if the reintroduction scheme is correct, a greatly increased activity of West Nile virus in Africa should be followed by an epidemic occurrence of West Nile fever in Europe in the next few years.

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