

of the Gulf of Mexico, where millions of neotropical migratory birds make landfall each year and spend the winter months. Beginning in 2000, a joint effort by the Universidad Autónoma de Yucatán and Colorado State University blood-sampled and tested migratory and resident birds in Yucatán State, Mexico. The following year, the Smithsonian Institution also began sampling birds on the Yucatán Peninsula. Further south in the Lacandón Forest of Chiapas State, a joint federal Mexico-United States study evaluated blood from about 200 resident domestic animals sampled in July of 2001. From these, a single seropositive cow (*Bos* sp.) with a PRNT₅₀ for WNV of 1:80 and a PRNT₅₀ for SLEV of 1:20, was considered a probable case of WNV infection (12). However, these authors cautioned against concluding that WNV had reached southern Mexico. They reasoned that a major range extension should be confirmed by a second detection of infection. Also, no evidence of WNV transmission had been detected at that time in the nearby Yucatán Peninsula (9, 13). The Chiapas study demonstrated serologic evidence for infections due to uncharacterized flaviviruses which could have resulted in cross-reaction with WNV. Secondary flavivirus infections are notorious for causing elevated heterologous flavivirus titers (14).

Spread of West Nile virus 2002–2004

In 2002, WNV continued to spread in the Caribbean Basin. Guadeloupe (French West Indies) reported numerous subclinical infections in horses and chickens, determined serologically by neutralization (15). In July 2002, 10.4% of the healthy horses in four locations were positive and by January 2003, 61.6% had become positive in these locations. The absence of reported neurologic disease in these horses is mysterious. Subsequent surveillance in 2003 and 2004 failed to detect any transmission (16).

In the Dominican Republic on the Greater Antillean island of Hispaniola, a University of Kansas study team

FIGURE 1. Countries of Latin America and the Caribbean with reported activity for West Nile virus (in black) between 2001 and 2004, including Mexico, Belize, Guatemala, El Salvador, Cuba, Bahamas, Cayman Islands, Jamaica, Dominican Republic, Puerto Rico (United States), Guadeloupe (French West Indies), Trinidad and Tobago, and Colombia



sampled blood and tissues from resident birds captured in November, 2002, for museum collections (17). Five birds of 33 (15.2%) from the Parque Nacional Los Haitises on the northeast coast tested positive for WNV antibodies by neutralization and a specific inhibition-ELISA test. A follow-up study in March, 2003, yielded 12 more WNV-seropositive birds of 58 (20.7%) at the Parque Nacional Monte Cristi in northwest Dominican Republic, along the border with Haiti (18). Positive Dominican bird species included *Phaenicophilus palmarum* ($n = 4$), two each of *Ploceus cucullatus*, *Saurothera longirostris*, *Loxigilla violacea* and *Turdus plumbeus*, and one each of five other species.

Evidence of WNV infection was confirmed in Mexico as of July, 2002. Seropositive horses were reported from six states (Chihuahua, Coahuila,

Tamaulipas, Veracruz, Tabasco and Yucatán) (19–21). Seropositive birds were rare and were first detected in the early winter months of 2003 (13, 22).

Mexican authorities began widespread serosurveys in horses and birds in 2003 and found many seropositive horses in 22 states (J. Mendez, personal communication, 4 Feb 2004), with no human cases in 2003 and six human cases (three with encephalitis) in northern Mexico in 2004 (23). The first Mexican isolate came from a dead captive common raven (*Corvus corax*) in Tabasco State (southeast Mexico) in May, 2003 (21). Additional isolates from dead birds were obtained in northwest Mexico later in 2003 and 2004. Phylogenetic analysis of the prM-E region of the WNV genome isolated from the raven in Tabasco linked it to central United States strains from 2002, but revealed slightly greater genetic variation than previous

reports for North American WNV strains (21). Two of the 9 nucleotide mutations resulted in amino acid changes, and one of these altered a glycosylation site within the envelope (E) protein. Virulence testing of plaque-purified subcultures of this isolate revealed variants with reduced virulence in mice (24). Similar observations had been made with a Texas 2002 isolate (25).

The widespread WNV seropositivity among horses observed in Mexico in 2003 was also present in the Central American republics of El Salvador and Guatemala (26, M.E. Morales-Betoulle et al., manuscript in preparation). However, anecdotal reports of fatal or life-threatening neurologic disease in Mexican and Central American horses have rarely been confirmed as due to WNV. One encephalitic horse diagnosed with WNV infection was reported from Belize, with onset October 31, 2003. Interestingly, 2 000 birds sampled in Belize earlier in 2003 and another 2 000 in 2002 all tested negative for WNV antibodies (27).

West Nile virus activity continued in the eastern Caribbean region in 2003. In the Bahamas, a human case of WNND was diagnosed with onset in July, 2003 (28). In early 2004, two seropositive *Turdus plumbeus* (of 734 birds sampled) were detected in Guantanamo Bay Naval Base at the eastern point of Cuba, and in eastern Puerto Rico, one *Coereba flaveola* (of 1 200 birds sampled) was seropositive, probably reflecting transmission in 2003 (29). Three neutralizing antibody- and IgM-positive, healthy horses were also reported in eastern Puerto Rico in May, 2004, and two others were found in central Puerto Rico in July, 2004 (A. Diaz et al., manuscript in preparation). Mosquitoes collected from the locations where seropositive horses resided tested negative for WNV infection. Four seropositive horses from the Havana region and three human WNND cases in central Cuba were announced in January, 2005 (G. Kouri, personal communication, 2 February 2005), reflecting transmission in 2004.

In the fall of 2004, 8 resident unvaccinated horses (of 200 sampled) and 2 domestic Muscovy ducks (of 40 resi-

dent birds sampled) were seropositive for WNV in Trinidad (28; R. Salas, personal communication, 17 November 2005), and 12 seropositive equines (of 130 sampled) were reported in northern Colombia (30). These reports mark the first evidence of WNV activity in South American ecosystems (the island of Trinidad is located within sight of the South American mainland off the coast of Venezuela). Efforts to detect WNV-specific antibodies in resident and migrant birds in Brazil in 2002 and 2003 were unsuccessful (31). With the incursion of WNV into northern South America in 2004, it becomes the only zoonotic flavivirus to have been identified in six continents.

DISCUSSION

The failure of efforts to isolate the virus or detect genomic RNA from WNV in Latin America and the Caribbean (with a few exceptions in Mexico) is perplexing and underscores the concern that serologic evidence for WNV activity is at best indirect. Flaviviruses are notorious for their close antigenic relationships and serologic cross-reactivity (10). In spite of strong serologic evidence from cross-neutralization testing against known flaviviruses from the region, the possibility of misdiagnosis due to cross-reaction with an as yet unrecognized "WN-like" virus still exists. In fact, some of the serologic results classified as due to "undifferentiated flavivirus infection" can best be explained by the existence of such a virus. The recent discovery of two strains of WN-like virus in central Europe lends credence to this concern (32). These two WN-like viruses were both identified serologically as WNV, but genetically they are equidistant from both currently recognized WNV lineages and each other and may represent newly discovered WNV lineages or new WN-like flaviviruses.

Another concern is the strong emphasis placed by several research groups on serologic surveillance of migratory birds (9, 13, 22, 29, 31). These studies consume large quantities of

valuable resources, yet are unlikely to provide significant results. Given the recent intense transmission of WNV during the summers in temperate North America, the capture of WN-seropositive avian survivors either during migration or on the wintering grounds is to be expected because many of these birds normally migrate to neotropical winter territories where they probably continue to circulate antibodies derived from a WNV infection acquired on their North American breeding grounds. Some studies claim that seropositive migrants are evidence that birds could carry WNV long distances. Unfortunately, although plausible, this conclusion is not valid for two reasons. First, the possibility that WN-seropositive migratory birds were in fact infected locally cannot be disproved. Second, long-distance migration by a healthy, antibody-circulating bird does not indicate that a viremic bird could make the same long-distance flight. More data are needed to support such a hypothesis. However, the observation of infectious WNV at high titers in tissues of convalescent migratory birds (e.g., Killdeer, *Charadrius vociferus*) more than one week post-infection and the demonstration of oral infection in raptors would suggest that recently infected birds that recover from viremia, migrate, and then fall prey to a raptor may still introduce WNV into new distant ecosystems if the raptor becomes infected and circulates sufficient virus in its blood to infect mosquitoes (33).

The most pressing concern regarding the reports of WNV in Latin America and the Caribbean is the absence of data on the disease burden in people, horses or birds. Widespread resistance to virulent strains of WNV in Latin American and Caribbean vertebrates (including people) seems highly unlikely. However, the selection of resistant WNV strains is plausible. If migrating birds are indeed the major mechanism for southward dispersal of WNV, then one could imagine a scenario in which birds infected with highly virulent strains become too sick to migrate, while birds infected with avirulent strains make the long flights

across seas and deserts successfully, spreading avirulent WNV to new transmission foci along their migratory routes. More research is needed to evaluate this hypothesis, but if proven, this bodes well for the future of WNV epidemics in North America, as the avirulent strain might be reintroduced continually from the south by returning migratory birds. South American arboviruses have in fact been isolated from northward-bound birds during the spring migration in Louisiana (34). This scenario may also explain the apparent low virulence for SLEV in birds and horses in North and South America. In fact, South American strains of SLEV are also less viremogenic in birds than are North American strains, and less virulent in mice (35). Whether an avirulent bird and horse strain of WNV will also be less virulent for humans remains to be seen.

Saint Louis encephalitis virus may be responsible for considerable cross-reaction to WNV in serologic tests of serum from Latin America. The virus is expected to cross-react in about 5% of primary WNV infections of birds (36). However, in secondary infections, the proportion of samples that cross-react by PRNT is probably much

greater. Secondary flavivirus infections may explain the high rate of flavivirus antibody-positive serum samples in the Caribbean Basin countries that cannot be assigned to a specific infection (because of the presence of similar titers for multiple flaviviruses). Although rarely associated with disease in Latin America, SLEV infections are commonly reported. For example, in Chiapas, Mexico, 20 (10%) of 196 domestic animals (including three of five horses) were diagnosed as positive for SLEV-neutralizing antibodies by PRNT (12). The known range of distribution for SLEV was expanded through the efforts to detect WNV in the Caribbean Basin. For example, two SLE-seropositive birds reported in Puerto Rico provide the first evidence of SLEV activity from that Caribbean location (9, 29).

CONCLUSION AND RECOMMENDATIONS

Although WNV has yet to present a serious disease threat in Latin America and the Caribbean Basin, an outbreak may be pending. The first major outbreak (with >100 human cases of

WNND) in the United States was delayed until 2002, three years after initial detection of the virus in 1999. Public health and veterinary authorities in Latin America and the Caribbean should remain vigilant for unusual clusters of severe disease cases. Dead birds (especially corvids) have been particularly useful for the early detection of WNV activity in North America (37). Corvids are less abundant in Latin America, and thus avian mortality may be less useful as a surveillance technique in this region (38). In countries where WNV has already been detected, surveillance efforts should be expanded. Surveillance guidelines for Latin American and Caribbean Basin countries are available (28, 39, 40).

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RESUMEN

La actividad del virus del Nilo occidental en América Latina y el Caribe

Objetivos. El virus del Nilo occidental (VNO, familia Flaviviridae, género *Flavivirus*) se ha propagado rápidamente por toda la cuenca del Caribe desde que se detectó por primera vez en 2001. En este informe se resumen nuestros conocimientos actuales acerca de la transmisión del VNO en zonas tropicales del continente americano.

Métodos. Revisamos todo lo que se ha publicado sobre el tema y consultamos a autoridades de salud clave para obtener datos inéditos.

Resultados. Las infecciones por el virus del Nilo occidental aparecieron por primera vez en seres humanos residentes de las Islas Caimán y de los Cayos de la Florida en 2001, y en pájaros de aspecto sano de los cuales se obtuvieron muestras a principios de 2002. En 2002 se encontraron pruebas serológicas de infección por el VNO en caballos, pollos y aves de corral no estabuladas oriundas de Guadalupe, la República Dominicana y la parte oriental de México. En 2003, el VNO se diseminó dentro de México y por la parte norte de Centroamérica y se encontraron pruebas serológicas en las Bahamas, Puerto Rico y Cuba. En 2004, las primeras pruebas serológicas de actividad vírica en ecosistemas sudamericanos se detectaron en septiembre y octubre en Colombia y Trinidad, donde se observaron anticuerpos neutralizantes contra el VNO en animales domésticos.

Conclusiones. Estos informes esporádicos de enfermedad equina, humana y aviar en América Latina y el Caribe son desconcertantes. Es necesario aislar las cepas para determinar si la atenuación del virus u otro factor explica la carga de enfermedad reducida en ecosistemas tropicales.

Palabras clave

Virus del Nilo occidental, América Latina, región del Caribe, arbovirus, vigilancia de la población, flavivirus.

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研究報告の概要	○クリミア・コンゴ出血熱—トルコ 2008年7月7日、トルコのブルサ、チャナッカレ、サムスン の病院で3名の患者がダニ媒介性疾患のクリミア・コンゴ出血熱で死亡し、この2ヶ月間での死亡者数は37名になった。 保健省はダニに注意するよう呼びかける声明を発表した。ダニに咬まれた場合は決して手でつぶさず、皮膚を保護し、医師にピンセットで注意深く取り除いてもらった後、ヨードで消毒することを推奨している。さらに、咬まれた人は10日間医学的観察を行い、発熱、頭痛、吐き気、嘔吐、下痢などの症状が現われた場合は、最寄りの病院を受診するよう、保健省の担当者は話している。 クリミア・コンゴ出血熱は主に動物に感染し、ヒツジや家畜に寄生するダニが、時折人にウイルスを感染させる。迅速に治療しないと出血によって死亡することもある。感染した人の血液や唾液を介して他の人にウイルスが伝播される可能性がある。感染地域はアフリカ、アジア、ヨーロッパの一部だが、近年トルコの気候が温暖になっていることから、ダニの数が増えてより多くの人が感染するようになっていると保健当局では話している。				使用上の注意記載状況・ その他参考事項等
	合成血-LR「日赤」 照射合成血-LR「日赤」 血液を介するウイルス、 細菌、原虫等の感染 vCJD等の伝播のリスク				
報告企業の意見		今後の対応			
2008年7月7日、トルコの病院で3名の患者がダニ媒介性疾患のクリミア・コンゴ出血熱で死亡し、この2ヶ月間での死亡者数は37名になったとの報告である。		日本赤十字社では、輸血感染症対策として問診時に海外渡航歴の有無を確認し、帰国(入国)後4週間は献血不適としている。今後も引き続き、新たな病原体による感染症の発生状況等に関する情報の収集に努める。			

