the etiology of dengue-associated relative bradycardia.

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References
1. Guzman MG, Kouri G. Dengue: an update. Lancet Infect Dis. 2002;2:33–42.
2. Fisher D. To the vector borne….mosquito-transmitted diseases in Singapore. Singapore Med J. 2005;46:596.
3. Wilder-Smith A, Earnest A, Paton NI. Use of simple laboratory features to distinguish the early stage of severe acute respiratory syndrome from dengue fever. Clin Infect Dis. 2004;39:1818–23.
4. Guzman MG, Kouri G. Dengue diagnosis, advances and challenges. Int J Cardiol. 1998;64:31–6.
5. Blatteis CM, Li S, Li Z, Feleder C, Perlk V. Cytokines, PGE2 and endotoxic fever: a reassessment. Prostaglandins Other Lipid Mediat. 2005;76:1–18.
6. Fink J, Gu F, Vasudevan SG. Role of T cells, cytokines and antibody in dengue fever and dengue haemorrhagic fever. Rev Med Virol. 2006;16:263–75.

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West Nile Virus, Venezuela

To the Editor: West Nile virus (WNV; genus Flavivirus; family Flaviviridae) has been perpetuating in North America since 1999 (1). However, its status as a self-perpetuating pathogen in South America remains uncertain. Infected horses and birds have been reported in various Caribbean Islands, Mexico, and northern Central America (2,3). In South America, isolated reports of infected dead-end hosts (horses) have come from northern Colombia and Argentina, but they lack evidence for infection in avian amplifying hosts (4,5). We report serologic evidence of establishment of WNV in South America.

Serum samples from birds and horses from 33 locations in Venezuela (Online Appendix Table, available from http://www.cdc.gov/EID/content/13/4/651-appT.htm) were screened for immunoglobulin G (IgG) antibodies against WNV antigen by ELISA (6) and confirmed by plaque reduction neutralization test (PRNT) as previously described (7). The flavivirus generating the IgG response was identified by using the following criteria: 90% inhibition of virus in serum diluted at least 1:40 and 4-fold greater neutralizing antibody titer compared with closely related flaviviruses. IgG antibody against flavivirus was detected by ELISA in 14 of 576 resident birds, including 5 Turdus leucomelas, 3 Gallus gallus (captive), 2 Campylorhamphus trochilirostris, and 1 each of Elaenia flavogaster, Coereba flaveola, Thraupis palmarum, and Anisognathus flavinucha.

WNV was confirmed as the etiologic agent of infection in 5 adult birds (3 T. leucomelas [pale-breasted thrush], 1 C. flaveola [bananquit], and 1 G. gallus [domestic chicken] with the earliest collection date in February 2006); virus neutralization...
Antibody against flavivirus was detected by ELISA in 141 of 791 horses, and 34 (4.3%) were confirmed positive for WNV infection by PRNT; viral titers ≥640 occurred in half of these horses. The earliest collection date for a WNV-positive horse was February 2004 and the most recent was May 2006. Specific WNV-reactive equine serum samples were distributed in valley regions (prevalence 1.3%), savannah grasslands (2.4%), the western region of Zulia (0.4%) and the Central Lake Basin (0.3%). A total of 46 (5.8%) equine serum samples were positive for neutralizing antibody to SLEV, and 8 (1.0%) samples were positive for neutralizing antibodies to ILHV. Forty-nine samples neutralized at least 2 of the 3 viruses and were classified as undetermined flaviviruses. Serum samples from 2 horses were negative in neutralization assays; 2 others were not tested because of insufficient sample volume.

Detection of WNV-infected resident birds provides strong evidence of the establishment, rather than importation, of WNV in South America. We hypothesize that ornithophilic mosquitoes (such as some Culex spp.), which are present in the area in consistently high numbers, acquired the virus through hematophagous feeding on recently infected, migrating birds. Once introduced to local mosquitoes, virus is amplified among susceptible resident birds fed upon by ornithophilic mosquitoes. This pattern allows perpetuation and subsequent establishment of virus in a continuous transmission cycle, as opposed to infection of dead-end hosts, e.g., horses. This is the first report of WNV infection in South American birds and definitive establishment of the virus in South America.

We observed varying WNV seroprevalence rates in birds and horses across regions in Venezuela (Figure). These differences reflect the focal and stochastic nature of arbovirus transmission, which depends upon many ecologic factors. One possible explanation for the greater seroprevalence in the central and eastern llanos (savannahs) and valley regions, compared with the coastal western region of Zulia State (p<0.0001, by Pearson’s χ² test) would be virus introduction by migrating birds by an eastern migration route.

Existence of several closely related flaviviruses in the American tropics (8–10) may convey cross-protection in animals (e.g., ILHV and SLEV) or humans (dengue viruses, yellow fever virus), thereby potentially diminishing disease caused by a newly introduced flavivirus such as WNV. Although ILHV infection has not been detected in Venezuela, this flavivirus is prevalent in Brazil, Peru, French Guyana, Trinidad, and Colombia. Our study demonstrated widespread distribution of ILHV in Venezuela. Other South American flaviviruses, such as Bussuquara, Cacipacore, and Iguape, and as yet undiscovered viruses may also circulate in Venezuela.

We encourage those involved in the public and animal health systems in Venezuela to consider zoonotic flaviviruses in the differential diagnoses of human and equine cases of encephalitis and to consider ecologic surveillance for zoonotic flaviviruses in mosquito and vertebrate host populations. We recommend monitoring blood and organ donations for flavivirus infections. Our study sheds light on flavivirus distribution in Venezuela. However, nothing else is known about the ecology of zoonotic flaviviruses in this country. Such knowledge will be essential for designing effective surveillance and control should these viruses be shown to cause human illnesses.
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References

1. Hayes EB, Komar N, Nasci RS, Montgomery SP, O’Leary DR. Campbell GL. Epidemiology and transmission dynamics of West Nile virus disease. Emerg Infect Dis. 2005;11:1167–73.
2. Komar N, Clark GG. West Nile virus activity in Latin America and the Caribbean. Rev Panam Salud Publica. 2006;19:112–7.
3. Morales-Betouille ME, Morales H, Blitvich BJ, Powers AM, Davis EA, Klein R, et al. West Nile virus in horses, Guatemala. Emerg Infect Dis. 2006;12:1038–9.
4. Mattar S, Edwards E, Laguardo J, Gonzalez M. Alvarez J, Komar N. West Nile virus antibodies in Colombian horses. Emerg Infect Dis. 2005;11:1497–8.
5. Morales MA, Barrandeguy M, Fabbri C, Garcia GB, Vissani A, Trono K, et al. West Nile virus isolation from equines in Argentina, 2006. Emerg Infect Dis. 2006;12:1559–61.
6. Ebel GD, Dupuis AP II, Nicholas D, Young D. Maffei J, Kramer LD. Detection by enzyme-linked immunosorbent assay of antibodies to West Nile virus in birds. Emerg Infect Dis. 2002;8:979–82.
7. Dupuis AP II, Marra PP, Kramer LD. Serologic evidence of West Nile virus transmission, Jamaica, West Indies. Emerg Infect Dis. 2003;9:860–3.
8. Figueiredo LT. The Brazilian flaviviruses. Microbes Infect. 2000;2:1643–9.
9. Kuno G, Chang GJ, Tsuchiya KR, harassment J, Powers AM, Davis EA, Klein R, et al. West Nile virus isolation from equines in Argentina, 2006. Emerg Infect Dis. 2005;11:1497–8.
10. Calisher CH, Monath TP. Karabatsos N, Cropp CB. Phylogeny of the genus Flavivirus. J Virol. 1998;72:73–83.

To the Editor: A 38-year-old French man with a history of chronic juvenile arthritis was referred to the Necker-Enfants Malades University hospital (Paris, France) with a dysenteric syndrome. The patient had returned the day before from a 1-month stay in Port-au-Prince, Haiti, where he spent most of his time in close contact with young children from an orphanage, most of whom had diarrhea. Clinical examination at admission showed fever (39°C), chills, diffuse abdominal pain, bloody diarrhea, and vomiting. The patient received ceftriaxone, which was stopped on day 4 because initial blood and stool cultures were negative for pathogens and clinical signs had completely resolved.

Ten days later, he reported the recurrence of diarrhea without fever. A novel stool culture grew Shigella sonnei. An extended-spectrum β-lactamase (ESBL) was detected by double-disc synergy test; the isolate was also resistant to aminoglycosides (except amikacin), tetracycline, and cotrimoxazole. The strain was susceptible to fluoroquinolones and fosfomycin. It also appeared susceptible to azithromycin (MIC 4 µg/mL), although azithromycin MIC for Shigella spp. should be interpreted with caution (I). The patient was successfully treated with azithromycin at a dose of 500 mg/day for 5 days. Azithromycin was preferred to fluoroquinolones to avoid the risk for tendinopathy because of the patient’s history of chronic juvenile arthritis and because this antimicrobial agent was shown to be effective in the treatment of shigellosis caused by multidrug-resistant strains (2).

To identify the molecular basis of this ESBL, a series of PCR primers

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### Online Appendix Table. Locations in Venezuela sampled for West Nile virus

| ID | State   | County, Location          | Regional ecosystem* | No. serum samples tested (no. positive)† |
|----|---------|---------------------------|--------------------|----------------------------------------|
| 4  | Aragua  | Guirardot, Portachuelos   | Coast              | 96 (0)                                 |
| 8  | Falcón  | Manuere, Guayabal         | Coast              | 7 (0)                                  |
| 25 | Zulia   | Francisco J. Pulgar       | Coast              | 6 (1)                                  |
| 26 | Zulia   | Mara                      | Coast              | 9 (1)                                  |
| 33 | Zulia   | La C. de Urdaneta         | Coast              | 12 (0)                                 |
| 27 | Zulia   | Maracaibo                 | Coast              | 327 (1)                                |
| 28 | Zulia   | Paez, Goajira             | Coast              | 6 (0)                                  |
| 2  | Apure   | Romulo Gallegos           | Llanos             | 25 (0)                                 |
| 10 | Guarico | Rivas                     | Llanos             | 43 (1)                                 |
| 11 | Guarico | Monagas, Altgr. Orituco   | Llanos             | 6 (3)                                  |
| 13 | Guarico | Ortiz, Zaraza             | Llanos             | 5 (0)                                  |
| 14 | Guarico | Infante, V. de la Pascua  | Llanos             | 18 (3)                                 |
| 15 | Guarico | Miranda, Guarico Dam      | Llanos             | 50 (3)                                 |
| 16 | Guarico | Miranda                   | Llanos             | 13 (5)                                 |
| 21 | Yaracuy | Bolivar                   | Valley             | 27 (2)                                 |
| 23 | Yaracuy | S. Felipe, Guaquira farm  | Valley             | 75 (0)                                 |
| 22 | Yaracuy | Veroes                    | Valley             | 24 (8)                                 |
| 5  | Barinas | Pedraza                   | Llanos             | 9 (4)                                  |
| 7  | Cojedes | Pao                       | Llanos             | 4 (0)                                  |
| 12 | Guarico | Las Mercedes              | Llanos             | 3 (0)                                  |
| 17 | Guarico | Monagas                   | Llanos             | 4 (0)                                  |
| 18 | Lara    | Urdaneta                  | Valley             | 1 (0)                                  |
| 19 | Lara    | Torres                    | Valley             | 3 (0)                                  |
| 20 | Lara    | Iribarren                 | Valley             | 3 (0)                                  |
| 24 | Yaracuy | Sucre                     | Valley             | 1 (0)                                  |
| 32 | Merida  | Libertador, Merida        | Mountain           | 5 (0)                                  |
| 3  | Aragua  | Girardot, Cata farm       | Coast              | 52 (0)                                 |
| 6  | Carabobo| Valencia, Lake            | Valley             | 16 (0)                                 |
| 29 | Zulia   | Paez, Sinamaica Lagoon    | Coast              | 142 (1)                                |
| 30 | Sucre   | Sucre, Cumana             | Coast              | 7 (1)                                  |
| 1  | Anzoategui| Miranda, F. El Tigre    | Llanos             | 56 (3)                                 |
| 9  | Falcón  | Iturriza, Cuare Reserve   | Coast              | 48 (0)                                 |
| 31 | Delta Amacuro| Tucupita            | Delta system       | 72 (0)                                 |

Total 576 (5) 791 (34)

* Llanos, savannah.
† Positive by 90% plaque reduction neutralization test.