DISCOVERY AND GEOGRAPHIC DISTRIBUTION OF VENEZUELAN ENCEPHALITIS VIRUS IN GUATEMALA, HONDURAS AND BRITISH HONDURAS, AND ITS POSSIBLE MOVEMENT IN CENTRAL AMERICA AND MEXICO

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Discovery and geographic distribution of Venezuelan encephalitis virus in Guatemala, Honduras and British Honduras, and its possible movement in Central America and Mexico*

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Introduction

For over 30 years, Venezuelan encephalitis (VE) virus and its diseases in humans and equines have been recognized in the countries of northern South America and Panama because the virus has repeatedly caused epidemics and equine epizootics (1). The discovery of VE virus antibody in serum from a human with neurologic sequelae in Campeche, Yucatan, Mexico during 1962 by Mexican investigators (2), the initial isolation of VE virus from mosquitoes in sentinel animals in the state of Veracruz, Mexico in 1963 (3), and the finding of the virus and antibody in the Everglades of southern Florida by United States Public Health Service, National Communicable Disease Center personnel during 1962-63 (4,5) have stimulated consideration that the virus might be spreading and increasing its geographic distribution in the Caribbean region.

To begin to evaluate this possibility, the geographic distribution of VE virus was investigated in Central America between its main locations in Panama and Mexico. This article describes studies in Guatemala, Honduras and British Honduras during 1965-68; studies carried out during 1967-68 by the Middle America Research Unit in Nicaragua and Costa Rica will be recorded elsewhere.

Methods

Exposure of sentinel hamsters and isolation of VE virus from their tissues were done as previously described (3,6).

VE virus antibody tests. Techniques of hemagglutination-inhibition (HI), neutralization (N) and complement-fixation (CF) tests were described elsewhere (7). N tests were done in cultures of primary chicken embryonic cell microcultures (8). VE virus, strain 63U2 from Mexico (3), was used as antigen in these tests.
Results

Since prior to these studies, there were no reported or recognized epidemics or equine epizootics of VE virus disease in Central America north of Panama, the presence of the virus in this region could not be established by study of cases during outbreaks. Therefore, investigations focused on likely ecologic habitats of the virus and were designed to isolate virus (by exposure of sentinel hamsters) and to detect antibodies (by HI, N and CF tests). Habitats considered most likely to contain VE virus were around marshes or within wet rain forests in tropical lowlands. Whenever possible, habitats near cities were studied to increase the potential importance of finding virus.

Isolations of VE virus by use of sentinel hamsters in Guatemala, Honduras and British Honduras. VE virus was isolated from sentinel hamsters exposed on the Atlantic lowlands at Puerto Barrios (1 strain), and Sayaxche (2 strains), Guatemala during August 1968, at Puerto Cortez, Honduras (5 strains) during July-October 1967, and at Belize, British Honduras (2 strains) during July-August 1967. On the Pacific lowlands of Guatemala, there were 5 isolations in the state of Retalhuleu and 2 near Avellana in the state of Santa Rosa.

These viruses were isolated by inoculation of hamster heart or brain suspensions into 1-4 day-old mice intracranially and subcutaneously, and were identified by CF and N tests. The 2 strains from Belize were reisolated in cultures of primary chicken embryonic cells. Selected strains from each location were studied with a VE virus antiserum which neutralized VE virus to a high LNI titer, but neutralized Mucambo and Pixuna viruses only to lower titers. From the Atlantic lowlands,
2 of 2 strains from Guatemala, 2 of 2 from British Honduras, and 3 of 5 from Honduras were studied in this manner and were shown to be VE virus rather than the closely related Mucambo or Pixuna viruses. From the Pacific lowlands, all 7 strains were similarly shown to be VE virus.

**Occurrence of antibodies to VE virus in humans in Guatemala, Honduras and British Honduras.** VE virus HI antibody was found in human sera from the Atlantic lowlands of Guatemala, Honduras and British Honduras during 1965-67, and from the Pacific lowlands of Guatemala during 1967-68. Some antibody titers were above 80 and, therefore, quite likely represented infection by VE virus and not by another group A arbovirus (9). Moreover, sera positive by HI were also positive by N test, and some were also positive by CF test suggesting recent or repeated infection.

**Geographic distribution of VE virus and its antibody in Guatemala, Honduras and British Honduras.** The focality of VE virus was manifested by isolations from sentinel hamsters. For example, in the Peten of Guatemala, the northern forested region, virus was abundant at Sayaxche, but not at Tikal, only 50-60 miles away. Both areas were forested, but Tikal was relatively dry because the ground was porous whereas Sayaxche was in a river basin and wet. In Honduras along the northern Atlantic coast, virus was isolated in a marsh habitat at the edge of Puerto Cortez, but not in the city proper or at nearby localities of San Pedro Sula-La Lima, Tela or La Ceiba, even though numerous sentinel hamsters were placed near water, marshes or forests and exposed for hundreds of hamster days. Similarly on the Pacific coast of Guatemala, virus was found in marsh habitats at La Chorrera, in a mangrove forest-swamp at Santa Sofia,
but not in nearby mangrove marshes at Champerico despite many hamster
days of exposure. In the Atlantic lowlands, sentinel hamster isolation
rates/100 hamster-days were 1.2 at Puerto Barrios and 1.7 at Sayaxche,
Guatemala, 0.6 at Puerto Cortez, Honduras, and 0.3 at Belize, British
Honduras. In the Pacific lowlands of Guatemala, they were 0.7 at La
Chorrera swamp, 23.5 at Santa Sofia and 0.7 at Avellana. The extra-
ordinarily high rate in the relatively small mangrove forest-swamp at
Santa Sofia is a striking example of focal virus activity. Focality
of VE virus was also suggested by antibody tests of human sera in the
Peten of Guatemala, but because people have moved from one area to
another in this country in recent years, these data by no means afford
proof of virus focality.

Although seasonal variation in VE virus prevalences was expected
from the known effects of wet and dry seasons on mosquito populations,
no such conclusions could be drawn from these sentinel hamster isolations,
because no one location was studied during both wet and dry seasons of
one year.
Discussion

VE virus was isolated by exposure of sentinel hamsters along the Atlantic lowlands of Guatemala, Honduras and British Honduras during July-October 1967-68 and on the Pacific lowlands of Guatemala during July-August 1968. It was found in wet marshy or forested habitats, both near cities and in remote areas. The presence of VE virus antibody in humans as early as August 1965 on the Atlantic lowlands and August 1967 on the Pacific lowlands of Guatemala probably established the presence of virus in these regions prior to those dates. Unfortunately, antibodies alone are not unequivocal evidence of the existence of VE virus since other group A arboviruses, such as Mayaro or Mucambo and Pixuna viruses in the VE complex produce antibodies that react with VE virus (9, 10). However, subsequent isolations of VE virus from the same regions where antibodies were found strongly suggested that at least some of the antibodies were specific for VE virus, and thus indicated the existence of the virus on the Atlantic lowlands of northern Central America prior to 1965.

These findings, together with those as yet unreported, of VE virus isolations by use of sentinel hamsters in the Atlantic lowlands of Nicaragua and Pacific lowlands of Costa Rica (11) suggest that this virus is now throughout Central America from Panama to Mexico at least along the Atlantic coast. Obviously, it is only speculative whether a) the virus has recently been transported from its previously known locations in Venezuela, Colombia, and Panama to Florida and through Central America to Mexico possibly by ships or airplanes (carrying infected mosquitoes or humans) or by infected, migrating birds, or b) the virus has been present in Central America, Mexico and Florida for many years,
centuries or longer and has now just been discovered, because arbovirus investigations were carried out in appropriate habitats. The geographic distribution of VE virus at least on the Atlantic coast of Central America from Panama to Mexico, supports the latter possibility. Also, the finding of virus in remote areas not limited to port regions in Central America and Mexico favors its presence for many years, or introduction by migrating birds rather than recent introduction by ships or airplanes. Unfortunately, age distributions of mosquito-borne virus antibodies cannot usually be interpreted to time spread of a virus to a locality. For example, antibodies may be limited to people over 20, not because virus was last there 20 or more years ago, but because those are the people who are outside, working in areas where there are infected mosquitoes. Consequently, surveys for VE virus antibodies are not very useful in timing virus in different localities.

The possibility of VE virus movement in Central America and Mexico, therefore, remains theoretical. Possibly, it can be studied by monitoring edges of virus activity where there are no natural habitat barriers to virus spread. If virus moves out of small foci or if it is spreading northward along the Gulf coast of Mexico as suggested by the occurrence of the first recognized disease in Mexico, a horse epizootic at Tampico, Tamaulipas in 1966 (12), one should be able to detect such movement by regular, repeated exposures of sentinel hamsters and tests for antibodies in appropriate vertebrate hosts.
References


