

SEROSURVEY AND VIRUS ISOLATION FROM RODENTS TO DETERMINE  
THE HANTAAAN VIRUS PRESENCE IN THAILAND  
(1 OCT. '82 - 30 SEPT. '83)

Principal Investigators : Markpol Tingpalapong, DVM  
James W. LeDuc, MAJ, MSC, USAMRIID  
Michael R. Elwell, MAJ, VC  
George S. Ward, LTC, VC  
Pranee Hansukjariya, BSc.

OBJECTIVES :

1. To identify areas in Thailand where rodents have antibody for Hantaan virus.
2. To test human sera from areas with rodent infection to determine if there is serological evidence of human infection.
3. To isolate Hantaan virus from tissues of rodents in endemic areas.

BACKGROUND : Recent studies have found Hantaan virus to be the causative agent of Korean hemorrhagic fever (KHF), a disease syndrome of potential military significance in Korea and Manchuria and of potential significance in the USSR, the Balkans, parts of Western Europe and Scandinavia.<sup>1</sup> Evidence has recently been obtained in Seoul, Korea that urban *Rattus* also are chronically infected with Hantaan virus. Cases of KHF in man have now been linked to infected wild rats in urban Seoul and Osaka, Japan.<sup>2</sup> In addition, antibodies to Hantaan virus have been found in *Rattus* captured near the docks in Japan, Korea and United States.<sup>3</sup> Chronic infection of rats and international shipping provide a chain which may have disseminated this virus worldwide. Therefore the potential for this agent to cause human disease may be far greater and more widespread than is presently appreciated. Last year we reported that rodents trapped at the Klong Toey port in Bangkok had antibody to Hantaan virus. In this preliminary study, approximately 20% of the bandicoots (*B. indicus*) tested had high antibody titer to Hantaan. Natural infection with Hantaan virus has not been previously reported to occur in this species.

METHODS : Rodents were live trapped at each field site (Klong Toey, Sriracha, Bangpakong, Kanchanaburi). They were anesthetized with ether, species were identified and bled. Serum was separated and kept on wet ice until frozen at -70°C. Bandicoots were euthanized and samples of lung, spleen, kidney and urine were kept on wet ice until frozen at -70°C. Exceptions were bandicoots trapped at Klong Toey. Tissue samples from these rodents were frozen at the time of harvest. Serum and tissues were shipped on dry ice to USAMRIID for antibody titer determinations (FA and PRNT) and virus isolation and IFA staining of tissue for virus antigen (in progress).

Blood samples were obtained from residents living around the fields where bandicoots were trapped. These were also tested for antibody by IFA.

RESULTS : Rodent trapping has been completed in three port areas in Thailand : Klong Toey in Bangkok, an international shipping port, and in Sriracha and Bangpakong, 2 grain shipping ports on the north eastern Gulf of Thailand. The numbers of rodents with positive antibody titers to Hantaan virus are shown by species (Table 1). A high percentage of positive titers was discovered in bandicoots from Klong Toey port. Serum from bandicoots trapped previously in several areas in Thailand was tested to see if antibody to this Hantaan or Hantaan-like agent was present in rodents from other locations. A small sample of eight sera from Korat and nine from Nakorn Pathom were all negative. However 3 of 3 samples from bandicoots trapped in Kanchanaburi in 1980 had positive titers (1:512-2048). A follow-up trapping of bandicoots in the same location in Kanchanaburi was completed in 1983; a high percentage of bandicoots had positive titers to Hantaan. In addition, sera from people who live in the rural area where those bandicoots were trapped were tested for Hantaan antibody (Table 2). Although the sample was small, 10/30 (33%) had titers  $\geq$  1:32 (range 1:32 - 1:512). This is in contrast to sample of 70 persons living in Bangkok near the port where only 4(6%) had titers of  $\geq$  1:32. It is now planned to determine if there is any evidence of human illness associated with this infection in Kanchanaburi.

#### REFERENCES :

1. Lee, H.W., Lee, P.W., and Johnson, K.M., 1978. Isolation of the etiologic agent of Korea hemorrhagic fever. *J. Infect. Dis.*, 137:298-308.
2. Ho Wang Lee, Luck J. Baek and Karl M. Johnson : Isolation of Hantaan virus, the Etiologic Agent of Korean Hemorrhagic Fever, from Wild Urban Rata. In Press.
3. LeDuc, J.W., Smith, G.A., *et al.*, 1982. Letter to the editor. *NEJM* 307:624.

Table 1. Antibody to Hantaan virus in rodents in Thailand.

| Location             | Species           |                  |                      |
|----------------------|-------------------|------------------|----------------------|
|                      | <i>B. indicus</i> | <i>R. rattus</i> | <i>R. norvegicus</i> |
| Bangkok (Klong Toey) | 6/29 <sup>a</sup> | 0/55             | 0/54                 |
| Sriracha             | 0/0               | 0/24             | 7/16                 |
| Bangpakong           | 0/0               | 6/86             | 0/0                  |
| Kanchanaburi         | 6/21              | 0/3              | 0/0                  |
| Total                | 12/50(24%)        | 6/168(3.6%)      | 4/70(5.7%)           |

<sup>a</sup> Number with titers  $\geq$  1:32 over total tested.

Table 2. Antibody to Hantaan virus in people in Kanchanaburi, Thailand.

| Age             | # Positive/# tested (range) |
|-----------------|-----------------------------|
| 0 - 9           | 0/0                         |
| 10 - 19         | 5/9 (1:32-512)              |
| 20 - 29         | 1/4 (1:32)                  |
| 30 - 39         | 1/8 (1:512)                 |
| 40 <sup>+</sup> | 3/12 (1:32)                 |
| Total           | 10/30                       |