

Title Virus Diseases of Americans in Southeast Asia

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Objective: To determine the etiology and study the epidemiology of viral illnesses in US military and civilian personnel stationed in Southeast Asia.

Description: Through liason with dispensaries and hospitals in Thailand specimens for virologic and serologic diagnoses were obtained.

Studies on fevers of unknown origin occurring in US military personnel in South Vietnam are being done through the cooperation of the US Army Medical Research Team (WRAIR) Vietnam and the medical staffs of the 93rd Evacuation Hospital and the 8th Field Hospital. Clinical and epidemiologic data and serum specimens were collected in these hospitals and the serum shipped to SMRL for virus isolation, and serologic testing for arboviruses (HI test) leptospirosis, (hemolytic test) and scrub typhus (FA test).

Progress:

Death due to Hemorrhagic Fever in an American Child.

Previous to this report no fully documented case of dengue shock syndrome had been observed in a caucasian in Thailand. The high incidence of dengue HF in Thai children and the absence of cases in caucasians had led to theories that genetic or nutritional differences caused the apparent difference in susceptibility. Recent evidence indicates that cases of dengue shock syndrome in Thai children are associated with a secondary antibody response to dengue infection. This report presents a fatal case of dengue HF in an American child with an immunologic response indicative of a secondary dengue infection.

A 16 month old male child born in Thailand of American parents had always lived in known dengue endemic areas of Thailand. The patient developed fever on 13 June 1966 and was brought to a medical facility on 16 June because of lethargy. Examination on 16 June revealed a fever (104°F), an erythematous circumoral rash and a palpable liver 2 cm. below the right costal margin. Antipyretics and tetracycline therapy were prescribed. On 17 June he appeared improved, total leucocyte count was 4600/mm<sup>3</sup> and the hematocrit was 37%. On 18 June he was admitted to the hospital because of lethargy, vomiting, and low fluid intake.

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Physical examination on admission revealed slight dehydration and lethargy, pulse 100/min, respirations 40/min, temperature 98.8°F. The lungs were clear and no cardiac abnormalities were noted. The liver was enlarged to 4 cm. below the costal margin. Scattered petechiae were seen on the lower extremities. Neurological examination was negative. Admission leucocyte count was 7,600/mm<sup>3</sup>, with 42% polymorphonuclear cells, 56% lymphocytes and 2% monocytes. Hemoglobin concentration was 16 gm%, erythrocyte count was 6.7 million/mm<sup>3</sup>.

Intravenous fluid therapy with 5% glucose in normal saline was started and erythromycin and dexamethasone were given. The patient suddenly expired 16 hours after admission.

At autopsy, scattered petechiae were present on the lower extremities. The pleural cavities contained about 200 ml. of serous fluid on each side and the peritoneal cavity contained 300 ml. of serous fluid. The liver extended 4 cm. below the costal margin and patchy capsular hemorrhages were present on the anterior surface. The stomach and intestines contained "coffee ground" fluid. Numerous scattered mucosal petechiae were present in the small and large intestine.

Microscopic examination of the liver revealed patchy hepatic necrosis characterized by hyalinization of hepatic and Kupffer cells. Focal perivascular hemorrhages were seen in sections of gastrointestinal tract. The spleen and lymph nodes showed lymphoid depletion and reticuloendothelial cell hyperplasia. Adrenal cortex revealed focal degeneration, and occasional cytolysis of the cortical cells, particularly of the zona glomerulosa and fasciculata.

No virus was isolated from heart blood obtained at death, 5 days after onset of fever. Antibody titers to the 4 dengue serotypes and Japanese encephalitis virus are listed in Table 36. Neutralizing (N) antibody titers showed broad cross reactivity within the dengue group. Sucrose density gradient ultracentrifugation of the serum (Table 37) revealed the presence of a small amount of 19S, mercaptoethanol sensitive antibody in fraction 4, while the majority of the antibody was in the 7S zone, fractions 6-9, and was mercaptoethanol resistant.

The 3 day febrile period followed by lethargy, increasing hepatomegaly, petechial rash, and hemoconcentration is typical of severe dengue hemorrhagic fever. The findings at autopsy of serous effusions, patchy capsular hemorrhages of the liver, petechiae of gastro-intestinal tract, focal hepatic necrosis, lymphoid depletion with associated reticuloendothelial cell hyperplasia of spleen, and lymphnodes and adrenal cortical damage have been described as characteristic findings after death due to dengue hemorrhagic fever. The clinical, and pathological findings, however, are not pathognomonic.

Neutralizing antibody titers following a first (primary) dengue infection are relatively low titered and do not exhibit extensive cross reactions with the other serotypes. Therefore, the high neutralizing antibody titers to all 4 dengue virus serotypes found in this case are indicative of a second infection with a dengue virus. The HI titer of 1:5120 on the 5th day after onset of illness is also evidence of a second infection with a dengue virus. Primary dengue infections have little or no HI antibody this early in the course of the illness.

The presence of 19S HI antibody establishes a recent dengue infection since 19S antibody persists only 3 to 6 weeks after onset of illness in dengue infections. The very high HI antibody titers in the 7S fractions of the sucrose density gradient on the fifth day after onset of illness are found in secondary dengue infections, but not in primary dengue infections.

Clinical, pathological, and serological findings provide strong evidence that the patient died of dengue hemorrhagic fever associated with a dengue virus infection. This case is of interest since it is the first documented case of severe dengue hemorrhagic fever in a caucasian of European descent, and suggests that factors other than genetics contribute to the apparent insusceptibility previously observed. If the "second infection hypothesis" of the pathogenesis of this disease is correct, the low incidence is due to the relatively small chance of foreigners living in Bangkok of contracting two dengue infections. The greater use of screens, repellants, and insecticides by foreigners, coupled with a limited time of residence in the country may be the most important factors.

FUO studies at the 93rd Evacuation Hospital. Between 1 April 1966 and 31 August 1966, an FUO study was carried out on patients admitted to the medical service of the 93rd Evacuation Hospital, Long Binh, with fever (over 101°F), chills, and headache, a negative malaria smear and in whom a specific diagnosis was not made within the first 24 hours. The results of this study were reported in detail in the Annual Progress Report, of the US Army Medical Research Team WRAIR (Vietnam), for the period ending 31 August 1966. The results of diagnostic studies on the 112 patients studied are summarized in table 38 for the purpose of comparison with data obtained in later studies.

A second study was begun in September 1966. All patients admitted to the medical service of the 93rd Evacuation Hospital between 1 September 66 and 15 February 67 with an unknown or uncertain diagnosis and a fever of 101° or higher on the morning following admission or a temperature elevation to 102° or greater during the first full hospital day were admitted to the study. Patients with malaria were admitted to the study if the diagnosis had not been confirmed by the morning following admission and if they met the established fever criteria.

Acute serum was drawn on all patients admitted to the study but convalescent serum was not obtained from those patients in whom a firm diagnosis was made by other means except in selected cases.

Eighty-six patients without firm diagnoses were lost to the study for administrative or operational reasons. Twenty-five paired samples were lost through breakage, spoilage, and other technical reasons. Five patients were evacuated from the area before convalescent serum could be obtained. The remaining 56 were discharged from the hospital before a second serum sample could be drawn. The clinical condition of the patients and the operational requirement for hospital beds prevented further delay in discharge and operational conditions prevented return of the patients.

Results are summarized in Tables 39, 40, and 41. A diagnosis was made on 69% of patients. Compared to the study done in April-August, dengue decreased markedly, chikungunya disease was not seen at all in the period September through February, and leptospirosis increased from 1% to 9% of cases.

The leptospirosis cases all occurred among troops in combat situations in III Corps area. Cases tended to cluster in units, 4 cases came from a single company with onset only a few days apart. Scrub typhus similarly occurred in troops on jungle operations and the cases in this study were contracted in III Corps.

FUO Study at the 8th Field Hospital. Sera were collected on 96 patients with clinical diagnosis of FUO at the 8th Field Hospital in October and November 1966. Malaria cases were not included in this study. Serologic studies indicated that of the 96 cases, 13 were dengue, 1 chikungunya, 1 Japanese encephalitis, 10 Leptospirosis and 8 scrub typhus. The leptospirosis and scrub typhus cases again occurred among combat troops on operations whereas the dengue infections were contracted in or near Nha Trang.

Distribution of arboviruses in Vietnam. In addition to the studies reported above, dengue was also serologically diagnosed in patients at the 3rd and 17th Field Hospital. If all are combined, a serologic diagnosis of dengue was made on 103 patients between March 1966 and February 1967. Cases occurred through the year with the largest number of cases (28) occurring in May and June corresponding to the peak incidence of hemorrhagic fever in Saigon.

Fourteen strains of dengue virus were isolated from acute phase sera. Of these, seven are dengue-1, two are dengue-2, three are dengue-4 and two are not yet identified. Dengue-1 strains came from cases originating in Saigon, Bien Hoa, Long Binh and Cu Chi. Dengue-2 strains came from Saigon and Ton San Nhut, and the dengue-4 strains came from Ton San Nhut and Bien Hoa. The unidentified strains came from Nha Trang and Duc My.

Chikungunya disease occurred in patients from Saigon, Long Binh, Bien Hoa and Tay Ninh. Only 13 cases were proven, and all occurred during the monsoon season (May-September).

Serologic confirmation of the diagnosis of Japanese encephalitis was obtained in 21 cases of acute central nervous system infections in US personnel. Five cases occurred in Da Nang in August 66, 4 occurred in Saigon or Ton San Nhut, 2 in Long Binh, 1 in Bien Hoa, 1 in Tay Ninh, 1 in Di An, 1 in Quang Duc province, 1 in An Khe, and 3 in Qui Nhon. Japanese encephalitis was also confirmed in 5 Vietnamese children in Quang Ngai where over 100 cases of encephalitis among Vietnamese children are reported annually.

Summary:

A fatal case of hemorrhagic fever in an American child was studied by serologic methods. Infection due to dengue virus and a secondary type antibody response was found.

Studies on fevers of unknown origin occurring in U.S. military personnel in South Vietnam indicated that dengue, leptospirosis, scrub typhus and chikungunya are significant causes of this syndrome. The relative prevalence of these infections varies with the season and with operational factors affecting exposure under combat conditions.

Dengue infections are common throughout III Corps in South Vietnam. In 1966 three dengue serotypes were identified as etiologic agents of FUOs in U.S. military personnel.

Japanese encephalitis was shown to cause central nervous system disease in widely separated regions of South Vietnam.