INTRODUCTION

Visceral leishmaniasis (VL) is an uncommon disease in Thailand. From 1960 to 1986, eight imported VL cases were reported (Laohapaibul and Siampakdi, 1960; Chutaputti et al., 1987; Suttinont et al., 1987). In 1996 the first non-imported VL case was reported in a 3-year-old girl from Surat Thani Province in southern Thailand. An investigation was unable to identify the source of infection (Bureau of Epidemiology, 1997). We investigated a second case of autochthonous VL in a 40-year-old Thai man who lived in Nan Province in northern Thailand and report here the characteristics of the case, the source of infection, the mode of transmission, and the likely vector.

MATERIALS AND METHODS

In July 2005, Chiang Mai University Hospital reported a case of VL to the Bureau of Epidemiology, Ministry of Public Health. We carried out the investigation in July and August 2005 in two settings: a rural village in Nan Province where the patient lived, and in the community in Bangkok where the patient had previously lived and worked. The neighborhood in Bangkok was populated by many non-Thais who had originated in known VL-endemic countries. The investigation focused on the section of the village in Nan Province and on households within a 100-meter radius of where the index patient lived in Bangkok.
We conducted active case and human source finding among villagers and proximal households in Bangkok. In the village, we defined a case as a person who lived in the village and had at least three of the following signs or symptoms: chronic fever, weight loss, anemia, or hepatosplenomegaly. We surveyed villagers door-to-door in the section of the village (approximately one-third of the entire village) where the index patient lived, collected blood for antibody testing and asked if they knew of other people in the village who had worked abroad and who had returned home prior to the onset of the patient’s illness, and if other foreigners had visited the village. In Bangkok we interviewed all the families living within a 100-meter radius of the index patient’s hut and asked about a history of chronic fever. No blood tests were done in Bangkok.

Environmental investigation

We also collected blood from domestic animals in the village, including dogs, cats, pigs and cows and tested their sera for Leishmania antibodies. We collected sand flies for speciation on two consecutives nights in the index case’s village between 5:00 PM and 5:00 AM. We used eight light traps, two Disney traps, four human-bait collectors, and a cow-bait net to increase the yield of sand flies. In Bangkok, we used four light traps to collect sand flies during a single night.

Laboratory testing

We tested the survey participants’ (human and animal) blood for Leishmania antibodies using the direct agglutination test (DAT) (KIT, Biomedical Research, the Netherlands). Positive samples were retested with PCR for Leishmania DNA (Fichoux et al, 1999).

RESULTS

Case history

The index patient was a 40-year-old Thai man. He had worked as a construction worker in several provinces, including Bangkok and Nan, between 1996 and 2004. He was addicted to amphetamines, opium and alcohol, but had no history of injecting drugs, blood transfusion, or travel abroad.

In December 2002, he had a sudden onset acute illness with intermittent fever and chills. The illness progressed over the next 15 months with intermittent fever, facial and extremity edema, abdominal pain, cough, bleeding gums, hematuria, melena, hemoptysis, hematemesis, anemia, and hepatosplenomegaly. He was first evaluated in March 2004. At that time, he had a chest radiograph that showed a large mediastinal mass. He was seen intermittently over the next 15 months until he was admitted at the Chiang Mai University Hospital, where VL was diagnosed by demonstration of Leishmania spp amastigotes and Leishmania donovani DNA from a bone marrow biopsy. He was treated with amphotericin B for 30 days. The parasite disappeared on a repeat bone marrow biopsy smear, and the patient’s condition and hematological features gradually improved. However, bleeding from the gums reoccurred two days after finishing the first course of treatment, and he was given a second 30-day course of amphotericin B. The time from onset of symptoms to first hospitalization was 15 months and to definitive diagnosis was 31 months.

Investigation

We interviewed 135 adults age 16-80 years old in the patient’s village and collected blood from 131 of them. None of them met our case definition for VL, and all sera were negative. One villager had returned from Libya in 2003, several months after the patient’s onset of disease; his serum also tested negative by DAT. We interviewed the heads of households of 151 families in Bangkok, 135 were Thai, 14 Nepalees and two other nationalities. No family members reported having a chronic fever.
We collected blood from 41 dogs, five cats, six pigs, and three cows in the village. All the pigs and dogs were negative by DAT, but one of five cats and all three cows were positive with antibody titers of 1:100 and 1:100-1:200, respectively. We checked for possible cross reactivity with other protozoan diseases that could possibly react with DAT by testing sera for Babesia, Trypanosoma, Anaplasma and Theileria spp and tested these with DAT, using bovine positive and negative controls. All these sera were negative. We attempted to confirm the presence of Leishmania DNA by PCR, but repeat tests from the seropositive animals were negative. We also collected blood from two rats found near the patient’s worksite in Bangkok, they tested negative by DAT.

Of 118 sand flies collected in the village, approximately 85% were Sergentomyia gemmae, 15% S. barraudi and two were Phlebotomus stantoni. Only three sand flies were collected from the neighborhood in Bangkok; all were Sergentomyia spp.

DISCUSSION

The clinical signs and symptoms of the index case (prolonged intermittent fever, anemia and hepatosplenomegaly) were compatible with VL. His diagnosis was confirmed by both microscopic visualization of Leishmania amastigotes on bone marrow and by a positive test for L. donovani by PCR. Remarkable weight loss was not observed, possibly due to edema and a low baseline body mass index, presumably due to his past alcoholism and amphetamine use. Because VL is a rare disease in Thailand, clinicians’ unfamiliarity with the disease may account for the delay in his diagnosis and appropriate treatment. Duration from first hospitalization until diagnosis for previous reported Thai VL cases ranged from two months to one year (Laohapaibul and Siampakdi, 1960; Suttinont et al, 1987; Chutaputti et al, 1987; Bureau of Epidemiology, 1997; Thisyakorn et al, 1999). For this case the delay was even longer. In India, where VL is endemic, the mean delay from the onset to diagnosis is 7.7 months (Sundar et al, 1991).

We could not identify the source of infection for the patient. The lack of evidence for another human reservoir may be due to the time lag between exposure and diagnosis, and frequent movement of the index case. The patient has never been outside Thailand. The existence of potentially competent sand fly vectors in Thailand (Apiwathanasorn et al, 1989) suggests that sand fly bites are more likely to have resulted in transmission to the index patient than human-to-human parenteral transmission. Nonetheless, asymptomatic humans can be a source of infection (Costa et al, 2002), and the patient’s history of methamphetamine use, which is typically smoked rather than injected in Thailand, could make him an unreliable historian.

Antibody evidence in cows and a cat were found, however, this can neither prove nor disprove these were a source for human infection. Domestic animals can be reservoirs of Leishmania infection (Abranches et al, 1998). We did not detect parasite DNA from the blood samples of those animals. However, the antibody findings highlighted the possibility that the asymptomatic infection can be harbored in domestic animals and that disease may be transmitted to humans if a vector is present in the environment. DAT is recommended to screen for VL in humans and dogs (Abdel-Hameed et al, 1989; Cardoso et al, 2004). Its sensitivity and specificity for testing VL in human and dog sera are more than 90% (Oskam et al, 1996; Boelaert et al, 2004). Though efficacy of the test in other species is uncertain, we found DAT could differentiate VL infection from other protozoan infections in cow serum.

We found that the large majority of sand fly species in our surveys were Sergentomyia.
Only two of 121 flies were P. stantoni. To our knowledge, none of these sand fly species have previously been shown to be vectors of leishmaniasis. Vector competency and species of sand flies vary by geographic setting. Several Phlebotomus spp (P. alexandri, P. chinensis, P. longiductus, P. sichuanesis and P. smernovi) are known to be competent vectors of human leishmaniasis in China (Zhang and Leng, 1997), while P. perniciosus was found to be a vector of VL in Italy (Ascione et al, 1996). Previous sand fly surveys in Thailand by Apiwathanasorn et al (1989) demonstrated the existence of a potential sand fly vector, P. argentipes, which has been shown to be a competent VL vector in India. We could find no reference to P. stantoni, which was found in our village surveys, being a competent vector. Some experts suggest that sand flies can change their blood meal targets from animals to humans, hence, any sand fly species that feeds on mammalian blood may act as a vector for Leishmania (personal observation). We suggest that further sand fly surveys and studies designed to demonstrate vector competence of sand fly species in Thailand are needed.

This is the second non-imported VL case in Thailand. We believe that the infection most likely occurred as a result of the bite of an infected sand fly. No new cases or human reservoirs were found in the study areas. However, there was serological evidence of, but not active infection with, VL in domestic animals in the index patient’s home village in Nan Province. Thus far, the vector for VL transmission for this case has not yet been identified, quite possibly due to our limited sandfly survey during the investigation. As part of our investigation we recommended sandfly control in the index case’s village on a regular basis, sentinel surveillance of sandflies and animal reservoirs in the village, and strengthened surveillance for human cases in Nan Province and elsewhere. Since the symptoms can be similar to other diseases and the diagnosis is rather difficult, education of the medical community is needed to increase awareness for early detection of cases. For the public, avoiding unnecessary exposure to sandflies by not sleeping outside a bednet should be practiced. Much remains to be learned about the ecology, range and transmission potential of this serious disease in Thailand.

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REFERENCES


