Bartonella spp. Infections, Thailand

To the Editor: Bartonella are fastidious hemotropic gram-negative bacteria with a worldwide distribution. In Thailand, Bartonella species have been demonstrated in mammalian hosts, including rodents, cats and dogs, and in potential vectors, including fleas (1–4). However, data on human infection have been limited to case reports (5,6) and 1 seroprevalence survey, which found a 5.5% prevalence of past B. henselae infection (7). No studies have systematically assessed the frequency, clinical characteristics, or epidemiology of human Bartonella infections in Thailand.

We conducted a prospective study to determine causes of acute febrile illness in 4 community hospitals, 2 in Chiang Rai (northern Thailand) and 2 in Khon Kaen (northeastern Thailand). We enrolled patients ≥7 years of age with a temperature >38°C who were brought to study hospitals for treatment from February 4, 2002, through March 28, 2003. Patients were excluded if they had a history of fever for ≥2 weeks or an infection that could be diagnosed clinically. Acute-phase serum samples were collected at the time of enrollment and convalescent-phase serum samples 3–5 weeks later. We enrolled nonfebrile control patients ≥14 years of age who had noninfectious conditions; acute-phase serum samples were collected. Clinical information was abstracted from patient charts. Nurses conducted physical examinations and personal interviews to collect information on patients’ demographic characteristics, exposures to animals, and outdoor activities.

Serum samples were tested for immunoglobulin (Ig) G antibodies to Bartonella spp. by immunofluorescent antibody assay at the Bartonella Laboratory of the Centers for Disease Control and Prevention, Fort Collins, CO, USA. Strains used for antigen production were: B. elizabethae (F9251), B. henselae (Houston-1), B. quintana (Fuller), and B. vinsonii subsp. vinsonii (Baker). Homologous hyperimmune serum specimens were produced in BALB/c mice as previously described (8). Bartonella infection was considered confirmed in febrile patients who had a ≥4-fold rise in IgG antibody titers and a convalescent-phase titer >64. Probable infection was defined as 1) a 4-fold antibody titer rise but convalescent-phase titers of 64, or 2) high and stable titers (≥512 in acute-phase and convalescent-phase serum samples), or 3) acute-phase titer ≥512 with a ≥4-fold titer fall. Paired serum samples from febrile patients were also tested for serologic evidence of other common causes of febrile illness in Southeast Asia.

Febrile patients with acute-phase and convalescent-phase IgG antibody titers <128 were considered not to have Bartonella infection; we compared demographic and clinical characteristics of these patients to Bartonella-infected patients. To evaluate potential risk factors, we compared Bartonella-infected case-patients ≥14 years of age without serologic evidence of other infections (n = 20) to nonfebrile controls with IgG to Bartonella <128 (n = 70). Age adjusted odds ratios (AORs) with 95% confidence intervals (CIs) were calculated.

Serologic testing was completed on paired serum samples for 336 (46%) of 732 febrile patients enrolled; 92 (27%) had serologically confirmed (50) or probable (42) Bartonella infections. Thirty-five (38%) of these 92 had serologic evidence of infection with another pathogen. The remaining 57 Bartonella-infected case-patients (34 confirmed, 23 probable) had a median age of 19 years (range 7–72 years); 65% were males, 47% were students, and 35% were rice farmers. Common clinical characteristics of Bartonella-infected patients included myalgias (83%), chills (79%), and headache (77%). Thirty (60%) patients had anemia (hemoglobin level <13 mg/dL); 18 (32%) had a hemoglobin level <12 mg/dL, and 4 (7%) had <11 mg/dL. When compared with 193 febrile patients without Bartonella infection, the 57 Bartonella-infected patients were similar in age and sex but were more likely to be rice farmers and were more likely to have leukocytosis (Table). Compared with the 70 nonfebrile controls, Bartonella-infected case-patients were more likely to report tick exposure (32% vs. 7.9%; AOR = 5.6, 95% CI 1.5–21) and outdoor activities (55% vs. 31%; AOR = 2.7, 95% CI 1.0–7.4) during the 2 weeks before...
illness onset. Prevalence of reported rat exposure and animal ownership (cats, dogs, pigs, cows, or buffaloes) was similar among case-patients and controls.

We describe the frequency and clinical characteristics of acute Bartonella infection among febrile patients in Thailand. Over 25% of patients with undifferentiated febrile illness had serologic evidence of Bartonella infection (including 15% serologically confirmed). Our findings indicate that Bartonella infections may be common and underrecognized causes of acute febrile illness in rural Thailand. Although our results are limited by lack of culture confirmation, we used conservative case definitions for serologic diagnosis and therefore believe that most cases represent true Bartonella infections. The common clinical features of anemia and leukocytosis and the frequent tick exposure and outdoor activity are consistent with known features of Bartonella infections and lend support to serologic findings. Because of the potential for serologic cross-reactivity between Bartonella species, we did not attempt species identification. The case-control study was therefore limited by grouping case-patients that were likely infected with different Bartonella species for which risk factors may differ. Such studies could lead to meaningful recommendations for prevention and control of Bartonella infections. Additional epidemiologic and transmission studies are needed to improve understanding of risk factors, identify key animal reservoirs and vectors, and ascertain transmission dynamics.

Acknowledgments

We are grateful for the contributions of the many study collaborators, especially K. Limpakarnjanarat, S. Thamthitiwat, P. Mock, U. Siangphoe, P. Srisaengchay, P. Sawatwong, and A. Nisalak; the dedicated study staff; and all volunteer study participants.

Saithip Bhengsri, Henry C. Baggett, Leonardo F. Peruski Jr, Christina Morway, Ying Bai, Tamara L. Fisk, Anusorn Sridhiasrdr, Susan A. Maloney, Scott F. Dowell, and Michael Kosoy

Author affiliations: Thailand Ministry of Public Health–US Centers for Disease Control and Prevention Collaboration, Nonthaburi, Thailand (S. Bhengsri, H.C. Baggett, L.F. Peruski Jr, T.L. Fisk, S.A. Maloney); Thailand Ministry of Public Health, Nonthaburi (A. Sridhiasrdr); Centers for Disease Control and Prevention, Atlanta, Georgia, USA (S.F. Dowell); and Centers for Disease Control and Prevention, Fort Collins, Colorado, USA (C. Morway, Y. Bai, M. Kosoy).

DOI: 10.3201/eid1604.090699

References

1. Maruyama S, Sakai T, Morita Y, Tanaka S, Kabeya H, Boonmar S, et al. Prevalence of Bartonella species and 16s rRNA gene types of Bartonella henselae from domestic cats in Thailand. Am J Trop Med Hyg. 2001;65:783–7.
2. Parola P, Sanogo OY, Ledrhusnee K, Zeaier Z, Chauvancy G, Gonzalez JP, et al. Identification of Rickettsia spp. and Bartonella spp. in fleas from the Thai-Myanmar border. Ann N Y Acad Sci. 2003;990:173–81. DOI: 10.1111/j.1749-6632.2003.tb07359.x
3. Castle KT, Kosoy M, Lerdthusnee K, Phelan L, Bai Y, Gage KL, et al. Prevalence and diversity of Bartonella in rodents of northern Thailand: a comparison with Bartonella in rodents from southern China. Am J Trop Med Hyg. 2004;70: 429–33.

4. Sukawat J, Xuejie Y, Hancock SJ, Hegarty BC, Nilsamrong P, Breitschwerdt EB. Serologic and molecular evidence of coinfection with multiple vector-borne pathogens in dogs from Thailand. J Vet Intern Med. 2001;15:453–62. DOI: 10.1892/0891-6640(2001)015<0453:SAMEOC>2.3.CO;2

5. Kosoy M, Morway C, Sheff KW, Bai Y, Colborn J, Chalcraft L, et al. Bartonella tamiiae sp. nov., a newly recognized pathogen isolated from three human patients from Thailand. J Clin Microbiol. 2008;46:772–5. DOI: 10.1128/JCM.02120-07

6. Paitoonpong L, Chitsomkasem A, Chantrakooppungool S, Kanjanahareutai S, Tribuddharat C, Sriefungfung S. Bartonella henselae: first reported isolate in a human in Thailand. Southeast Asian J Trop Med Public Health. 2008;39:123–9.

7. Maruyama S, Boonmar S, Morita Y, Sakai T, Tanaka S, Yamaguchi F, et al. Seroprevalence of Bartonella henselae and Toxoplasma gondii among healthy individuals in Thailand. J Vet Med Sci. 2000;62:635–7. DOI: 10.1292/jvms.62.635

8. Kosoy MY, Regnery RL, Tzianabos T, Marston EL, Jones DC, Green D, et al. Distribution, diversity, and host specificity of Bartonella in rodents from the southeastern United States. Am J Trop Med Hyg. 1997;57:578–88.

Address for correspondence: Saithip Bhengsri, International Emerging Infections Program, Thailand Ministry of Public Health–US Centers for Disease Control and Prevention Collaboration, 3rd Floor, Bldg 7, Department of Disease Control, Ministry of Public Health, Nonthaburi, 11000, Thailand; email: saithipb@th.cdc.gov

**Cholera Outbreak, Laos, 2007**

**To the Editor:** Cholera is a major public health problem in countries where access to safe water and adequate sanitation cannot be guaranteed for all. *Vibrio cholerae* serogroups O1 and O139 are the causative agents of cholera (1). One of the most powerful virulence factors in this organism is cholera toxin encoded by the ctxAB gene, located on the CTX prophage. *V. cholerae* O1 is classified into 2 biotypes, classical and El Tor. The El Tor type of *V. cholerae* O1 is responsible for the ongoing seventh worldwide pandemic of cholera (2). The sequence of ctxB of a certain strain has been believed to correspond to its biotype; that is, a biotype classical strain has classical type ctxB, and a biotype El Tor strain has El Tor type ctxB. However, recent research studies suggest that novel types of *V. cholerae* O1, hybrid strains, and altered El Tor or El Tor variant strains (1,3) are emerging. Altered El Tor or El Tor variant strains are biotype El Tor but produce classical cholera toxin (3,4). Recent reports suggest that this type of *V. cholerae* O1 is spreading to many areas of the world (5).

In December 2007–January 2008, a cholera outbreak occurred in Xekong Province in southeastern Laos, in the Mekong Basin. The first case of the outbreak was detected on December 23, 2007. The outbreak spread to 10 villages and lasted through January 2008. Specifically, in the Thateng District, 117 cases occurred and 2 deaths were reported. The sources of the outbreak were suspected to be regularly used water. In October 2007, 2 months before the outbreak, 3 sporadic cases of *V. cholerae* infection had been identified in Vientiane (the capital city) and Xaignabouri Province in north-central and northwestern Laos, respectively. The outbreak investigation in the Xekong Province identified no linkage between these sporadic cases and the outbreak cases.

In this study, we analyzed 18 *V. cholerae* isolates obtained in 2007: 3 were from patients with sporadic cases, and 15 were from the Xekong outbreak (13 from patients and 2 from water samples). All the isolates were serotype O1 Ogawa and biotype El Tor, but their ctxB types were classical, according to the method previously described (6). This finding indicates that they were the type of altered El Tor.

We used pulsed-field gel electrophoresis (PFGE) to investigate relationships between the isolates according to the PulseNet protocol (7). All 18 isolates from the sporadic cases and the outbreak in 2007 displayed profiles indistinguishable from each other (Figure). We also compared 2 additional *V. cholerae* O1 isolates, 1 from a patient in Vientiane in 1998 and another from a patient in Louangphabang in 2000 (Figure). The profiles of the isolates obtained in 1998 and 2000 clearly differed from those obtained in 2007. These results indicate that all isolates from sporadic and outbreak cases in 2007 were likely from the same source of contamination, although extensive epidemiologic investigation did not identify any common source.

Nguyen et al. characterized the isolates from a cholera outbreak in Vietnam from late 2007 to early 2008 (8). Their report suggests that the isolates from the outbreaks in Vietnam and Laos shared the same elements of the CTX prophage. Our study suggests a common source for the strains of sporadic cases in Vientiane and Xaignabouri Province in October 2007 and those of the outbreak in Xekong Province in December 2007. Molecular typing suggests that a novel clone of *V. cholerae* O1 is being disseminated along the Mekong Basin. However, no epidemiologic association has been identified so far. Thus, a more extensive regionwide surveillance system is needed to identify and control *V. cholerae* infection in Laos and neighboring countries.