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Short Report: Detection of Rickettsioses and Q fever in Sri Lanka

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Abstract. Current serological evidence suggests the presence of scrub typhus and spotted fever group (SFG) rickettsiosis in Sri Lanka. Our objective was to identify rickettsial agents/Q fever as aetiological causes for patients who were presumed having rickettsioses by the presence of an eschar or a rash. Sera from patients with unknown origin fever from Matara were tested by immunofluorescence for SFG rickettsial antigens, typhus group rickettsiae, *Orientia tsutsugamushi*, and *Coxiella burnetii* antigens. Thirteen (7.3%) of the patients presented with a rash, 11 (6.1%) had an inoculation eschar, and 16 patients recalled a tick or flea bite. We found that 25 (14%) patients had scrub typhus, 6 (3%) SFG rickettsioses, 3 (1.6%) acute Q fever, 3 (1.6%) murine typhus, and 3 (1.6%) were infected by *Rickettsia felis.* In addition to already described scrub and murine typhus, we found that *R. felis* and *C. burnetii* infections should be considered in Sri Lanka.

INTRODUCTION

Rickettsial diseases are caused by obligate intracellular Gram-negative bacteria of the genus *Rickettsia*.[1] Four groups of diseases are called rickettsioses: 1) diseases caused by bacteria of the genus *Rickettsia*, including the spotted fever group (SFG) and the typhus group; 2) scrub typhus caused by *Orientia tsutsugamushi*; 3) human ehrlichiosis and anaplasmosis; and 4) the ubiquitous Q fever caused by *Coxiella burnetii*, which was, however, recently removed from the Rickettsiales.[1] Current serological evidence suggests the presence of scrub typhus and SFG rickettsiosis in Sri Lanka.[2] Q fever is a worldwide zoonosis with many acute and chronic manifestations, and cattle, sheep, and goats are the most important reservoirs for *C. burnetii.*[3] Experimental and epidemiological evidence clearly show that contaminated aerosols are the main source of *C. burnetii* contamination in humans.[4] The objective of this study was to analyze a large number of patients with persistent fever from southern Sri Lanka and to identify, based on serological assays, the causative agents.

THE STUDY

After ethical approval from the Ethical Review Committee, Faculty of Medicine, University of Peradeniya, Sri Lanka, patients in Medical Wards of District General Hospital, Matara, Sri Lanka who were presumed having rickettsioses by the presence of an eschar or a rash were further investigated retrospectively for *Rickettsia* and Q fever infection from March until September 2009. Serum samples from these patients were sent for examination at Unité des Rickettsies, Marseille, France. All sera were tested by immunofluorescence for spotted fever group (SFG) rickettsial antigens (*Rickettsia conorii conorii*, *Rickettsia india*, *Rickettsia japonica*, *Rickettsia felis*, *Rickettsia honei*, and *Rickettsia heilongjiangensis*), typhus group rickettsiae (*Rickettsia typhi*), *Orientia tsutsugamushi* (Gilliam, Kuroki, Sennetsu, and Kawasaki serotypes) and *C. burnetii* phase I and II antigens, as previously described.[4,5] Serum with a phase II immunoglobulin G (IgG) titer ≥ 200 and a phase II IgM titer ≥ 50 was predictive for acute Q fever. This serologic criterion was used to define acute Q fever. If a phase I IgG titer was ≥ 800, chronic Q fever was suspected. The presence of low levels of IgG antibodies was considered to be evidence of past infection.[4] Immunofluorescence was considered positive for *Rickettsia* spp. or *O. tsutsugamushi* infection when there was a 4-fold rise in the antibody titer or a single antibody titer of IgG ≥ 1/128 combined with an IgM titer ≥ 1/64 against one or more antigens of the tested species.[5]

Overall, we tested 178 patients with fever. An acute serum sample was received for all the patients and for 54 of them we also received a convalescent-phase serum sample. Out of these patients a rickettsial or Q fever etiology was found in 37 (20%) and 3 patients (1.6%) respectively. The duration of fever at presentation at the hospital ranged from 3 to 15 days (38.2 to 40.1°C). Thirteen patients (7.3%) presented with a rash and 11 (6.1%) had an inoculation eschar. Ten patients recalled a tick bite, and 6 patients recalled a flea bite. Scrub typhus was diagnosed in 25 patients (14%) (Table 1). Both the acute and the convalescent-phase serum samples of seven patients were positive for *O. tsutsugamushi,* and 5 patients had a negative acute serum sample and a positive convalescent-phase serum sample for *O. tsutsugamushi*. Among the remaining 13 patients positive for scrub typhus, we received only one serum sample. Twenty-three scrub-typhus-positive patients (92%) were infected with *O. tsutsugamushi* serotype Gilliam, 1 (4%) with serotype Kuroki, and 1 (4%) with serotype Kawasaki. The mean ± SD for the duration of fever for these patients with scrub typhus was 10 ± 5 days. Eight patients (30%) presented with skin eschars, 6 (24%) reported a tick bite, and 3 patients (12%) reported a rash. The SFG was diagnosed in 6 patients (3%). The mean ± SD duration for fever with SFG was 5 ± 2 days. One patient (16%) presented with a rash, and one recalled a tick bite. *Rickettsia felis* was diagnosed in 3 patients, with mean ± SD duration for fever of 4 ± 1 days. One patient with *R. felis* infection recalled a tick bite. Murine typhus was diagnosed in 3 patients (1.6%), with mean ± SD duration for fever of 4 ± 1 days. None of the patients infected by *R. typhi* presented with a rash. Acute Q fever was diagnosed in 3 patients (1.6%). All 3 patients had an IFA positive for *C. burnetii.* For only one of these patients did we receive a convalescent-phase serum sample, which was also positive and confirmed the diagnosis. The mean ± SD

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TABLE 1
Clinical information about patients

	No. of patients (%)	Days of fever*	Rash (%)	Tick bite (%)	Eschar (%)
Orientia tsutsugamushi	25 (14%)	10 ± 5	3 (12%)	0	8 (30%)
Spotted fever group	6 (3%)	5 ± 2	1 (16%)	1 (16%)	0
Rickettsia felis	3 (1.6%)	4 ± 1	0	1 (16%)	0
Rickettsia typhi	3 (1.6%)	4 ± 1	0	0	0
Coxiella burnetii	3 (1.6%)	9 ± 2	0	0	0

* Duration of fever before arriving at the hospital and before receiving any treatment.

duration for fever was 9 ± 2 days, and one patient presented with a rash. Past evidence of Q fever infection was detected for one patient.

DISCUSSION

Our results clearly indicate that SFG rickettsiosis, *R. felis* infection, scrub typhus, murine typhus, and Q fever are present in southern Sri Lanka. Our immunofluorescence assays for the detection of SFG, *C. burnetii*, *O. tsutsugamushi*, *R. typhi*, and *R. felis* are routinely used and have been previously validated as a reference for rickettsiosis and other arthropod bacterial diseases.[4,5] Scrub typhus, murine typhus, and SFG rickettsioses have been previously documented in Sri Lanka.[2,6] In our study, the presence of an inoculation eschar in patients with scrub typhus was less common (30%) than in the study of Premaratna and others[2] (89%), which could be caused by patient selection criteria, whereas the prevalence of rashes was similar in both studies. None of our patients with murine typhus presented with a rash. In cases of murine typhus, the rash is nonspecific, and its prevalence differs, as 20% of patients from Thailand presented with a rash, compared with 49% of patients from Texas, 80% of patients from Greece, and 62.5% of patients from Spain.[7] In Laos, 13% of patients with murine typhus presented with a skin rash, and the clinical profile of patients with murine typhus was similar to those with scrub typhus.[8] In a previous study in Sri Lanka, two patients with murine typhus infection presented with a rash.[6] Although SFG rickettsiosis are frequently identified as a cause of fever in Sri Lanka,[2,6] this is the first time that *R. felis* infection has been identified in this country. In Asia, *R. felis* infection has been previously described in Japan, Indonesia, Thailand, Afghanistan, Israel, Laos, Taiwan, and Lebanon.[9] In addition, we diagnosed Q fever infection in three patients. Q fever has been described worldwide, except in New Zealand.[4] Previous sero-epidemiological studies in animals revealed that *C. burnetii* is endemic in Sri Lanka. Kovacova and others[10] found that ~25% of goats and 15% of cattle exhibited antibodies against *C. burnetii*. Moreover, Sixl and others[11–13] found evidence of *C. burnetii* infection in cattle, goats, dogs, and crows from Sri Lanka. In a previous unpublished study, Q fever infection was found in 4% of febrile patients from the Gampaha district of Sri Lanka (Bailey MS and others, unpublished data).

In conclusion, our data can affect local clinical practice because in addition to scrub and murine typhus, we found that Q fever and *R. felis* infections are endemic in Sri Lanka and should be considered in the diagnosis of patients with fever. As a result, early empiric antibiotic therapy with doxycycline (200 mg/day) should be prescribed in any suspected rickettsiosis, before confirmation of the diagnosis.[14]

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