

Murine Typhus and Leptospirosis as Causes of Acute Undifferentiated Fever, Indonesia

M. Hussein Gasem,¹ Jiri F.P. Wagenaar,¹
Marga G.A. Goris, Mateus S. Adi,
Bambang B. Isbandrio, Rudy A. Hartskeerl,
Jean-Marc Rolain, Didier Raoult,
and Eric C.M. van Gorp

To investigate rickettsioses and leptospirosis among urban residents of Semarang, Indonesia, we tested the blood of 137 patients with fever. Evidence of *Rickettsia typhi*, the agent of murine typhus, was found in 9 patients. Another 9 patients showed inconclusive serologic results. Thirteen patients received a diagnosis of leptospirosis. No dual infections were detected.

Fever is one of the main reasons for seeking medical attention in Indonesia. The cause of fever usually remains obscure because of limited laboratory diagnostic facilities and expertise in performing laboratory confirmation. Favorable environmental conditions mean that both rickettsiosis and leptospirosis are considered endemic in Indonesia and may result in clinically indistinguishable cases of acute undifferentiated fever (AUF). Serosurveys conducted on Java, Sumatra, and islands in eastern Indonesia identified antibodies to *Rickettsia typhi* (murine typhus), to *Orientia tsutsugamushi* (scrub typhus), and to members of the spotted fever group rickettsia (SFGR) in healthy persons (1–3). In addition, several investigations reported leptospirosis as cause of AUF in Indonesia (4,5).

Murine typhus and leptospirosis are likely to share routes of transmission in an urban setting where rats are abundant. The main vector for *R. typhi* is the Asiatic rat flea (*Xenopsylla cheopsis*). Humans usually become infected when *R. typhi*-infected flea feces contaminates excoriated skin or is inhaled. Leptospirosis is mainly spread by rats and other small mammals, which shed the bacteria through their

urine into the environment. Humans are infected through mucous membranes, conjunctivae, or abraded skin.

The clinical features of mild leptospirosis and murine typhus are nonspecific. Generally, patients with murine typhus exhibit fever, headache, and a rash, although the latter is often absent. Renal failure, jaundice, and hemorrhages are the classic symptoms of severe leptospirosis; fever, headache, and myalgia may be the only exhibited symptoms of mild disease. Dual infections with murine typhus are reported to occur in Southeast Asia and may complicate treatment and clinical course (6,7). In this study, we attempted to find evidence for acute rickettsial disease, leptospirosis, and dual infections among patients with AUF in Indonesia, where risk factors for both diseases are present.

The Study

The study was based in Semarang, a large harbor city in central Java. Consecutive outpatients were recruited at 2 primary healthcare centers and hospitalized patients at a governmental referral center (Dr. Kariadi University Hospital, Department of Internal Medicine). All eligible AUF patients (≥ 5 years of age) were included who met the following criteria: fever $\geq 38^{\circ}\text{C}$ (central) for < 14 days with no apparent other disease. After informed consent was obtained, a blood sample was taken. A convalescent-phase sample was drawn after ≈ 14 days. The study was approved by the local medical ethical committee.

A specific microimmunofluorescent antibody (IFA) assay for *Rickettsia* spp. was performed in Marseille, France, by using whole-cell antigens of *O. tsutsugamushi*, *R. japonica*, *R. heilongjiangensis*, *R. slovacica*, *R. honei*, *R. conorii* subsp. *indica*, *Rickettsia* ATI, *R. helvetica*, *R. felis*, *R. typhi*, and *R. prowazekii*. The assay results were considered positive when 1) antibody titers were ≥ 256 for immunoglobulin (Ig) G and ≥ 64 for IgM, or 2) seroconversion was observed, or 3) a ≥ 4 -fold increase in titers between the acute-phase and the convalescent-phase serum specimen was detected. Serologic analysis for leptospirosis was performed in Semarang, Indonesia. Crosschecks and PCR were performed in Amsterdam, the Netherlands. Convalescent-phase samples were screened with the LeptoTek Dri Dot (bioMérieux, Marcy l'Etoile, France). All positive samples were tested by the microscopic agglutination test (MAT) and IgG ELISA (8).

Additionally, a real-time PCR specifically targeting the *secY* gene of pathogenic *Leptospira* spp. (9) was performed on all samples. For the MAT, a panel of 31 serovars was used containing 28 pathogenic and 3 nonpathogenic serovars. For patient samples tested by ELISA or MAT, a titer ≥ 320 on a single sample was considered positive; also considered positive were those samples that showed sero-

Author affiliations: Diponegoro University, Semarang, Indonesia (M.H. Gasem, M.S. Adi, B.B. Isbandrio); Slotervaart Hospital, Amsterdam, the Netherlands (J.F.P. Wagenaar, E.C.M. van Gorp); Royal Tropical Institute, Amsterdam (M.G.A. Goris, R.A. Hartskeerl); and Université de la Méditerranée, Marseille, France (J.M. Rolain, D. Raoult)

DOI: 10.3201/eid1506.081405

¹These authors equally contributed to this article.

conversion or a ≥ 4 -fold increase in titers between paired samples, as well as any patient sample with a positive PCR, irrespective of the serologic result. All samples were run in parallel.

From February 2005 through February 2006, 137 AUF patients were included: 67 hospitalized patients and 70 outpatients. A convalescent-phase sample was available for 106 (77%) patients. The main symptoms were headache (85%), myalgia (70%), nausea (64%), cough (44%), and abdominal pain (38%).

Murine typhus and leptospirosis were found to cause AUF in this clinical series (online Appendix Table, available from www.cdc.gov/EID/content/15/6/975-appT.htm). In total, 9 patients (7%) had evidence of an acute infection with *R. typhi*; none showed a rash. Murine typhus could be diagnosed in 6 (9%) of 67 hospitalized patients; 3 (4%) of 70 outpatients had acute murine typhus. Another 9 (7%) patients showed inconclusive *R. typhi* serologic results. One patient showed evidence of a past infection with *R. typhi* (IFA IgG/IgM titer 128/0 in both serum specimens). Evidence for acute infection with *O. tsutsugamushi* or SFGR was not found.

Leptospirosis was diagnosed in 13 (10%) of 137 patients; results for 2 of these patients were positive only by PCR. Eleven leptospirosis patients were recruited in the hospital; 2 patients were recruited outside the hospital. Consequently, the percentage of AUF caused by leptospirosis in hospitalized patients was 16% and in outpatients 3%. The most frequently identified serogroup by MAT was Bataviae (5 cases). No dual infections were detected; however, 3 (23%) leptospirosis patients showed titers in the *R. typhi* IFA assay.

Conclusions

We report that murine typhus and leptospirosis are important causes of AUF in Semarang, Indonesia. A previous study from rural Thailand identified both diseases in 2.8% and 36.9% of AUF cases, respectively (6,7). In Vientiane, the capital of Laos, *R. typhi* was reported to cause fever in 9.6% of investigated persons, results that closely resembling our data (10). Unfortunately, leptospirosis was not investigated.

In the present study, we expected leptospirosis to be a cause of AUF because the Dr. Kariadi University Hospital admits ≈ 50 severe cases each year. These cases were not included in the study because of the high clinical suspicion of leptospirosis on admission with jaundice, azotemia, and/or bleeding. A definite diagnosis of murine typhus and leptospirosis co-infections could not be made, but in 3 cases this scenario was plausible. We did not find evidence for scrub typhus, which we expected, because *O. tsutsugamushi* transmission occurs primarily in rural areas (11). Although SFGR have been reported in Southeast Asia and proof for

their presence in Indonesia is accumulating (2,12), these rickettsia were not identified as a cause of AUF in the present study.

From an epidemiologic point of view, Semarang, Indonesia, seems to encompass environmental circumstances that are prerequisites for *R. typhi* and leptospirosis transmission. Previous studies have shown that murine typhus is particularly prevalent in tropical port cities where rats are abundant (13,14). In the Indonesian urban situation, *R. rattus* and *R. norvegicus* rats are likely to be the main hosts harboring *R. typhi*-infected *X. cheopsis* fleas (12,15). These rats are also likely to be the maintenance hosts for pathogenic *Leptospira* spp. in Indonesia. In fact, the identified serogroups are commonly associated with rats.

Although serologic analysis might be hampered by cross-reactions and test sensitivity issues, we believe that our data are representative for the area. The chosen cut-off values are unlikely to cause false-positive results in a disease-endemic setting. In regard to leptospirosis serologic analysis, we used a wide panel for the MAT. This panel included serovars recommended by the World Health Organization and serovars that were previously isolated in Indonesia. Moreover, most serogroups were represented in our panel, and cross-reactions are likely to detect missing serovars.

Because of nonspecific clinical features, both diseases are difficult to diagnose on clinical grounds only. Misdiagnosis can lead to aberrant use of antimicrobial drugs and other pharmaceuticals. Therefore, rapid, cheap, and reliable diagnostic tests are needed to support clinical decision making.

Acknowledgments

We thank S.M.H. Faradz and personnel at the Center for Biomedical Research, Semarang, Indonesia, and the residents of the Department of Internal Medicine, Dr. Kariadi Hospital, Semarang, for their help and assistance during the study. We are also grateful to A.A. Ahmed for performing the leptospirosis PCR.

Dr Gasem is internist and acting head of Division of Infectious Disease, Department of Internal Medicine, Dr. Kariadi University Hospital. His research interests are typhoid fever and leptospirosis.

References

1. Richards AL, Soeatmadji DW, Widodo MA, Sardjono TW, Yanuwati B, Hernowati TE, et al. Seroepidemiologic evidence for murine and scrub typhus in Malang, Indonesia. *Am J Trop Med Hyg.* 1997;57:91–5.
2. Richards AL, Ratiwayanto S, Rahardjo E, Kelly DJ, Dasch GA, Fryauff DJ, et al. Serologic evidence of infection with ehrlichiae and spotted fever group rickettsiae among residents of Gag Island, Indonesia. *Am J Trop Med Hyg.* 2003;68:480–4.

3. Van Peenen PF, Koesharjono C, See R, Bourgeois AL, Irving GS. Antibodies against murine typhus in sera from Indonesians. *Trans R Soc Trop Med Hyg.* 1977;71:297–9. DOI: 10.1016/0035-9203-(77)90103-1
4. Light RH, Nasution R, Van Peenen PF. Leptospirosis in febrile hospital patients in Djakarta. *Southeast Asian J Trop Med Public Health.* 1971;2:493–5.
5. Laras K, Cao BV, Bounlu K, Nguyen TK, Olson JG, Thongchanh S, et al. The importance of leptospirosis in Southeast Asia. *Am J Trop Med Hyg.* 2002;67:278–86.
6. Ellis RD, Fukuda MM, McDaniel P, Welch K, Nisalak A, Murray CK, et al. Causes of fever in adults on the Thai-Myanmar border. *Am J Trop Med Hyg.* 2006;74:108–13.
7. Suttinont C, Losuwanaluk K, Niwatayakul K, Hoontrakul S, Intaranongpai W, Silpasakorn S, et al. Causes of acute, undifferentiated, febrile illness in rural Thailand: results of a prospective observational study. *Ann Trop Med Parasitol.* 2006;100:363–70. DOI: 10.1179/136485906X112158
8. Terpstra WJ, Ligthart GS, Schoone GJ. Serodiagnosis of human leptospirosis by enzyme-linked-immunosorbent-assay (ELISA). *Zentralbl Bakteriol A.* 1980;247:400–5.
9. Victoria B, Ahmed A, Zuerner RL, Ahmed N, Bulach DM, Quinteiro J, et al. Conservation of the S10-spc-alpha locus within otherwise highly plastic genomes provides phylogenetic insight into the genus *Leptospira*. *PLoS One.* 2008;3:e2752. DOI: 10.1371/journal.pone.0002752
10. Phongmany S, Rolain JM, Phetsouvanh R, Blacksell SD, Soukha-seum V, Rasachack B, et al. Rickettsial infections and fever, Vientiane, Laos. *Emerg Infect Dis.* 2006;12:256–62.
11. Watt G, Parola P. Scrub typhus and tropical rickettsioses. *Curr Opin Infect Dis.* 2003;16:429–36. DOI: 10.1097/00001432-200310000-00009
12. Ibrahim IN, Okabayashi T, Ristiyanto, Lestari EW, Yanase T, Muramatsu Y, et al. Serosurvey of wild rodents for rickettsioses (spotted fever, murine typhus and Q fever) in Java Island, Indonesia. *Eur J Epidemiol.* 1999;15:89–93. DOI: 10.1023/A:1007547721171
13. Richards AL, Rahardjo E, Rusjdi AF, Kelly DJ, Dasch GA, Church CJ, et al. Evidence of *Rickettsia typhi* and the potential for murine typhus in Jayapura, Irian Jaya, Indonesia. *Am J Trop Med Hyg.* 2002;66:431–4.
14. Dupont HT, Brouqui P, Faugere B, Raoult D. Prevalence of antibodies to *Coxiella burnetii*, *Rickettsia conorii*, and *Rickettsia typhi* in seven African countries. *Clin Infect Dis.* 1995;21:1126–33.
15. Jiang J, Soeatmadji DW, Henry KM, Ratiwayanto S, Bangs MJ, Richards AL. *Rickettsia felis* in *Xenopsylla cheopis*, Java, Indonesia. *Emerg Infect Dis.* 2006;12:1281–3.

Address for correspondence: Jiri F.P. Wagenaar, Department of Internal Medicine (9B), Slotervaart Hospital, Louwesweg 6, 1066 EC Amsterdam, the Netherlands; email: jfpwagenaar@hotmail.com

etymologia

Typhus

[ti' fəs]

From Greek τῆφος [*typhos*], meaning heavy stupor; also related to Greek *typhein*, to smoke. A disease known since antiquity, typhus has been described as follows: “A kind of continued fever, attended with great prostration of the nervous and vascular systems, with a tendency to putrefaction in the fluids and vitiation in the secretions; putrid fever. A genus of the order *Febres*, class *Pyrexia*, of Cullen’s nosology” (J. Thomas, 1885).

Today, typhus refers to any of a group of acute infections caused by rickettsiae and transmitted to persons by the bite of arthropods such as fleas and lice. Epidemic typhus, caused by *Rickettsia prowazekii*, is characterized by headache, high fever, chills, rash, and, in serious cases, by stupor or lack of awareness of reality. Outbreaks usually occur in crowded or unsanitary environments.

Source: Dorland’s illustrated medical dictionary, 31st ed. Philadelphia: Saunders; 2007; <http://www.merriam-webster.com>; Thomas J. A complete pronouncing medical dictionary. Philadelphia: JB Lippincott; 1885.

Appendix Table. Clinical and laboratory data of patients with rickettsioses and leptospirosis, Indonesia, February 2005–February 2006*

Disease/ patient no.	Age, y/sex	Month of illness onset	<i>Rickettsia typhi</i> IFA (IgG/IgM)		MAT		Leptospirosis PCR result	Clinical features and treatment
			Admission sample	Convalescent- phase sample	Highest titer pathogenic serovar	Putative infecting serogroup		
Murine typhus								
7	49/M	Feb	0/0	1,024/1,024	NA	NA	–	4-d fever, myalgia, headache, chills, cough, abdominal pain; Rx: ciprofloxacin
10	45/M	Mar	512/1,024	512/1,024	NA	NA	–	7-d fever, myalgia, headache, nausea, abdominal pain; Rx: ciprofloxacin
12	46/F	Mar	2,048/512	2,048/512	NA	NA	–	3-d fever, headache, chills, nausea, vomiting, hepatomegaly; Rx: ciprofloxacin
18	15/M	Apr	256/64	256/64	NA	NA	–	3-d fever, headache, sore throat, abdominal pain; Rx: cefadroxyl
24	46/F	Apr	128/512	1,024/1,024	NA	NA	–	7-d fever, headache, nausea, abdominal pain; Rx: ciprofloxacin
63	68/F	Nov	2,048/256	2,048/512	NA	NA	–	3-d fever, myalgia, headache, chills, nausea, dyspnea; Rx: ciprofloxacin
1107	13/F	Feb	0/32	1,024/512	NA	NA	–	2-d fever, headache, anorexia; Rx: cotrimoxazol
1112	25/M	Feb	256/64	NA	NA	NA	–	2-d fever, myalgia, headache, anorexia, sore throat, cough, abdominal pain, calf pain; Rx: amoxicillin
1123	8/F	Mar	0/64	512/0	NA	NA	–	1-d fever, headache, sore throat, nausea, vomiting, abdominal pain; Rx: amoxicillin
Possible murine typhus								
2	17/F	Feb	0/0	0/64	NA	NA	–	5-d fever, myalgia, headache, chills, cough sore throat; no antimicrobial drug
5	31/F	Feb	0/0	128/0	NA	NA	–	5-d fever, myalgia, headache, chills, nausea; no antimicrobial drug
53	22/F	Aug	128/32	128/64	NA	NA	–	4-d fever, myalgia, headache, nausea, vomiting, abdominal pain, diarrhea; no antimicrobial drug
62	17/M	Nov	128/64	128/64	NA	NA	–	4-d fever, myalgia, headache, chills, sore throat, nausea, vomiting, abdominal pain, petechiae; no antimicrobial drug
69	46/M	Dec	0/128	0/128	NA	NA	–	9-d fever, myalgia, headache, chills, nausea, abdominal pain; Rx: ciprofloxacin

Publisher: CDC; Journal: Emerging Infectious Diseases
Article Type: Dispatch; Volume: 15; Issue: 6; Year: 2009; Article ID: 08-1405
DOI: 10.3201/eid1506.081405; TOC Head: Dispatch

1104	14/M	Feb	128/64	128/64	NA	NA	–	1-d fever, headache; Rx: amoxicillin
1105	11/F	Feb	128/32	NA	NA	NA	–	2-d fever, myalgia, headache, nausea, vomiting, abdominal pain; Rx: amoxicillin
1113	70/F	Feb	0/0	0/64	NA	NA	–	1-d fever, myalgia, headache, sore throat, cough, calve pain; Rx: amoxicillin
1132	7/M	Mar	256/0	NA	NA	NA	NA†	3-d fever, headache, sore throat, nausea, vomiting, conjunctival suffusion, petechiae; Rx: amoxicillin
<hr/>								
Leptospirosis								
4	27/M	Feb	–	–	80†	Javanica	–	2-d fever, myalgia, headache, chills, nausea, vomiting, abdominal pain, calve pain, hepatomegaly; Rx: cefotaxime
14	63/M	Mar	128/0	128/0	1,280	Hebdomadis	–	3-d fever, myalgia, headache, nausea, vomiting, abdominal pain; Rx: ciprofloxacin
25	41/F	May	–	–	80†	Bataviae	–	2-d fever, myalgia, headache, chills, nausea, vomiting; no antimicrobial drug
29	49/M	May	–	–	160†	Bataviae	–	4-d fever, myalgia, headache, chills, diarrhea, conjunctival suffusion; Rx: ciprofloxacin and PP
32	40/M	May	–	–	320	Bataviae	–	5-d fever, myalgia, headache, nausea, vomiting, abdominal pain, petechiae; no antimicrobial drug
37	50/M	May	–	–	40†	NC	–	5-d fever, headache, chills, cough, nausea, vomiting, abdominal pain, diarrhea; Rx: PP
40	63/M	Jun	–	–	–	NC	+	2 d fever, myalgia, headache, nausea, vomiting, abdominal pain, calve pain; Rx: PP
48	16/M	Jun	–	–	640	Saxkoebing	–	2-d fever, myalgia, headache, chills, petechiae; no antimicrobial drug
1152	17/M	Mar	–	–	160†	Bataviae	–	2-d fever, myalgia, headache, chills, sore throat, cough, nausea; Rx: amoxicillin, tetracycline, cotrimoxazol
1182	58/F	Jan	–	–	320§	NC	+	2-d fever, myalgia, headache, cough; Rx: cotrimoxazol
<hr/>								
Possible co-infection								
33	38/M	May	1,024/0	1,024/0	320	Icterohemorrhagiae	–	5-d fever, myalgia, headache, nausea, abdominal pain, diarrhea, calve pain; Rx: cefotaxime

Publisher: CDC; Journal: Emerging Infectious Diseases
Article Type: Dispatch; Volume: 15; Issue: 6; Year: 2009; Article ID: 08-1405
DOI: 10.3201/eid1506.081405; TOC Head: Dispatch

43	39/F	Jul	0/0	0/256	640	Bataviae	+	5-d fever, myalgia, headache, chills, nausea, vomiting, jaundice, calve pain; Rx: PP
60	37/M	Oct	0/64	64/64	-	NC	+	5-d fever, myalgia, headache, nausea, abdominal pain; no antimicrobial drug

*IFA, microimmunofluorescent antibody; Ig, immunoglobulin; MAT, microscopic agglutination test; NA, not available; NC, not classifiable; PP, procaine penicillin; Rx, medical prescription.

†Patient seroconverted from negative (<20) to a positive titer by MAT or ELISA.

‡Insufficient sample to perform PCR; leptospirosis highly suspected in patient.

§IgG ELISA result; the highest titer by MAT was <40.