# Seroprevalence of Anti-Leptospira IgG and IgM Antibodies and Risk Assessment of Leptospirosis among Urban Poor Communities in Kuala Lumpur, Malaysia

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Abstract. Leptospirosis is a zoonotic bacterial disease caused by pathogenic species of the genus *Leptospira*. Disease incidence is known to be attributed to environmental and social conditions which promote the spread of reservoir hosts, primarily rodents. A well-being program was conducted to determine the seroprevalence and risk factors associated with leptospirosis in urban poor communities occupying low-cost flat accommodation and squatter settlements in the vicinity of Wilayah Persekutuan, Kuala Lumpur. Blood samples from a total of 532 volunteers were screened for the detection of IgG and IgM antibodies against leptospirosis using ELISA. Demographic data were collected for each participant through a questionnaire survey before blood collection. The overall seroprevalence was low (12.6%, n = 67/532; 95% CI: 9.9–15.7%), with 8.1% (n = 43/532) being seropositive for anti-*Leptospira* IgG, indicating previous infection, and 4.9% (n = 26/532) for anti-*Leptospira* IgM, indicating current infection. Two significant factors such as host age ( $P \le 0.01$ ) and knowledge of disease transmission (P = 0.017) significantly influenced the presence of anti-*Leptospira* IgM, whereas the detection of anti-IgG indicated the presence of clean drinking water sources (P = 0.043). Despite the low prevalence, the transmission of leptospirosis does occur among urban poor communities, suggesting the need for undertaking public awareness programs.

## INTRODUCTION

The cumulative growth of urbanization in Malaysia from 43.2% (1989) to 71% (2010) has resulted in a population increase from 27% in 1970 to 71% in 2010.<sup>1</sup> Many members of the population flock into the city to seek better lives, and such an increase in population density in the urban environment can impact on public health risks through the creation of dense infrastructures and inadequate sanitation, thereby facilitating the spread of communicable diseases. Urban poor is described as a condition whereby individuals or communities live in a city with the inability to cover costs for basic living needs. Urbanization, local and global migration from rural settlements to urban cities, together with increasing costs of living has contributed to a rise in urban poverty in Malaysia.<sup>2</sup> Urban health risks are also distributed unequally and largely among marginalized social groups, particularly those living in slum areas where up to 40% of urban population growth has occurred. Air-borne diseases, such as tuberculosis, are associated with overcrowding and inadequate ventilation, whereas water- and vector-borne diseases, such as leptospirosis, are linked to unsafe water storage and poor waste management.

Leptospirosis has been recognized globally as a zoonotic disease and a major cause of illness both in humans and animals and is caused by spirochete bacteria belonging to the genus *Leptospira*.<sup>3</sup> A total of 22 species has been described and classified according to DNA–DNA hybridization and phylogenetic analysis,<sup>4,5</sup> with more than 300 serovars based on agglutinating lipopolysaccharide antigens.<sup>6</sup> Human transmission is via direct contact with infected blood, tissues, organs, or urine of infected hosts or through indirect contact with contaminated fomites, soil, mud, fresh water, and rodent-infested habitats.<sup>7,8</sup> Transmission can also occur via direct penetration of *Leptospira* through the conjunctiva or surface epithelium.<sup>9</sup> The role of rats as a source of human infection was discovered in 1917 and recognized as a most important reservoir for infection as these rodents are abundant in most environments. Many clinical manifestations are observed with leptospirosis, ranging from asymptomatic and mild symptoms to a self-limited febrile and fulminant life-threatening one.<sup>7</sup>

Leptospirosis is highly prevalent and considered to be a reemerging disease in the Asia Pacific region. Malaysia is ranked in the top 20 countries, relative to high incidences of leptospirosis,<sup>10</sup> which increased sharply from 2,268 cases in 2011 to 8,291 in 2015.<sup>11</sup> The state of Wilayah Persekutuan in Kuala Lumpur is ranked with the highest number of outbreaks in the country, especially due to overcrowding, poverty, and poor sanitation in urban slum areas.<sup>12,13</sup> The present study targeted communities residing in the People's Housing Program (Program Perumahan Rakyat [PPR]) developed by the Ministry of Housing and Local Government Malaysia. Such a housing program was established following the demolition of squatter dwellings previously occupied by individuals within lower income groups and to fulfill their need for low-cost housing. Generally, urban housing comprises high-density flats equipped with basic facilities including clean water and sanitation. However, the situation is a far cry from reality as most PPR developments in Kuala Lumpur are vastly overcrowded with poor waste management.14

In view of the wide range of clinical manifestations shown by leptospirosis, severe cases are only detected when hospitalized. Therefore, a seroprevalence study was conducted for the first time in the urban poor community of Wilayah Persekutuan to identify risk factors associated with leptospirosis infection.

## METHODS

**Study population.** Using a well-being community program, an investigation of a targeted community within the state of Wilayah Persekutuan, Kuala Lumpur, was undertaken between

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October 2017 and March 2018 (Table 1, Figure 1). A minimum sample size of the population was calculated using a formula by Leedy and Ormrod<sup>15</sup> and based on earlier estimates of prevalence (12.6%) in Malaysia.<sup>16</sup> A total of 532 volunteers were successfully recruited, and each individual was given a set of questionnaires relating to sociodemographic factors, health status, environmental health, and awareness of leptospirosis. Consent forms were collected from each individual before collection of blood samples. Ethical clearance was obtained before commencing the study (reference number: BK-MIS-1117-E01).

**Sample collection.** A 5-mL sample of venous blood was drawn from each volunteer by medically trained personnel, and the samples were placed into BD Vacutainer SST<sup>M</sup> II Advance Plus (Fisher Scientific, Loughborough, Leicestershire, UK) blood collection tubes (each with a yellow top cap). Each tube was kept in an ice box and immediately transported to the Parasitology Laboratory, Institute of Biological Sciences, University of Malaya. Tubes were centrifuged using a benchtop centrifuge (Universal 320) (Hettich, Tuttlingen, Germany) at 1,500 × *g* for 10 minutes and the serum samples maintained at  $-20^{\circ}$ C until use.

Detection of immunoglobulin G and M antibodies to Leptospira spp. Seropositivity for Leptospira spp. infection was demonstrated by anti-Leptospira IgG and IgM antibodies using standard ELISA commercial kits (SERION ELISA classic, Institut Virion/Serion GmbH, Warburg, Germany). All reagents were maintained at room temperature for testing and sera allowed to thaw at the same temperature.

First, washing solutions were prepared by diluting the buffer concentrate (V1) 1:30 with distilled water to reach a final volume of 1,000 mL (V2). Before running the test, a rheumatoid factor (Rf) absorbent was used to dilute the buffer at a ratio of 1:4. A total of 200  $\mu$ L of Rf absorbent was added to 800  $\mu$ L of dilution buffer, and then in the case of IgM *Leptospira* kits, 10  $\mu$ L of each volunteer's sample was diluted with 1,000  $\mu$ L of Rf dilution buffer. For IgG *Leptospira* kits, 10  $\mu$ L of volunteer samples was diluted with 1,000  $\mu$ L of Rf dilution buffer (1:100). Following dilution and before pipetting into microtiter plates, samples were mixed thoroughly to ensure the solutions were homogenous.

Rheumatoid factors are autoantibodies mainly of the IgM class, which preferably bind to IgG immune complexes. The presence of nonspecific IgM antibodies (Rfs) can lead to false-positive results, and in addition, weak-binding pathogen-specific IgM antibodies may be displaced by stronger binding ones such as IgG, leading to false-negatives. Samples must, therefore, be treated with Rf absorbents before antibody detection. Sera must also be diluted with buffer during the assay because heterophilic antibodies in human serum/plasma are

capable of binding to both capture and detection antibodies and cross-linking can result in false-positives.

Apart from the blank substrate, each of 100 µL of diluted sample or ready-to-use controls were added to appropriate wells of the microtiter test strips and incubated at 37°C for 60 minutes in a moist chamber. Following incubation, wells were washed four times with 300 µL washing solution and 100 µL of the ready-to-use antihuman IgM conjugate (for Leptospira IgM) and antihuman IgG conjugate (for Leptospira IgG). Apart from the blank substrate, alkaline phosphatase conjugate was added to the wells and incubated again for 30 minutes. Following the second incubation, all wells were rinsed with the washing solution and 100 µL of ready-to-use p-nitrophenyl phosphate (pNPP) substrate was added, followed by incubation at 37°C for 30 minutes in a moist chamber. Finally, 100 µL of sodium hydroxide (stopping solution) was added to each well, and following gentle shaking of the microtiter plate, the enzyme-substrate reaction was stopped. Within 60 minutes, optical densities of samples against blank substrates were read at a wavelength of 405 nm (yellow water-soluble reaction products absorb light at this wavelength) against a background of 650 nm.

**Data analysis.** Seroprevalences of infection (%) are shown with 95% CIs as described by Rohlf and Sokal.<sup>17</sup> Seroprevalences were analyzed using maximum likelihood techniques based on log linear analysis of contingency tables using the software package SPSS (version 22) (IBM, Armonk, NY).

#### RESULTS

Seroprevalences of anti-*Leptospira* IgG and IgM within the poor communities. A total of 532 participants from 15 locations were involved in this study, and these locations included PPR Seri Anggerik (n = 53; 9.96%), Kampung Lembah Melewar (n = 7; 1.32%), PPR Desa Tun Razak (n = 53; 9.96%), Perumahan Awam Putra Ria (n = 31; 5.83%), PPR Intan Baiduri (n = 34; 6.39%), PPR Seri Cempaka (n = 66; 12.41%), PPR Sri Pantai (n = 46; 8.65%), Flat Perbadanan Kemajuan Negeri Selangor (PKNS) (n = 12; 2.26%), PPR Kampung Limau (n =41; 7.71%), Flat Vista Angkasa (n = 24; 4.51%), PPR Pantai Permai (n = 37; 6.95%), Kampung Sungai Penchala (n = 76; 14.29%), PPR Batu Muda (n = 8; 1.50%), PPR Kerinchi (n = 6; 1.13%), and PPR Pantai Ria (n = 38; 7.1%).

The overall seroprevalence of *Leptospira* infections among 532 participants was 12.6% (n = 67; 95% CI: 9.9–15.7%), with 8.1% (n = 43; 95% CI: 5.9–10.7%) seropositive to anti-*Leptospira* IgG and 4.9% (n = 26; 95% CI: 3.2–7.1%) seropositive to anti-*Leptospira* IgM. Only a total of 0.4% (n = 2; 95% CI: 0–1.4%) were positive to both anti-*Leptospira* IgG and IgM.

TABLE 1
Locations of urban poor communities with global positioning system (GPS) coordinates

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Locations	Coordinates	Locations	Coordinates
PPR Kerinchi	3.107149, 101.668984	Flat PKNS	3.112573, 101.663235
PPR Seri Anggerik	3.092198, 101.673858	PPR Kampung Limau	3.109022, 101.674798
Kampung Lembah Melewar	3.218245, 101.698487	Flat Vista Angkasa	3.113418, 101.661690
PPR Desa Tun Razak	3.078596, 101.717112	PPR Pantai Permai	3.108372, 101.668760
Perumahan Awam Putra Ria	3.123049, 101.676385	Kampung Sungai Penchala	3.162505, 101.625649
PPR Intan Baiduri	3.235292, 101.655946	PPR Batu Muda	3.208055, 101.682405
PPR Seri Cempaka	3.100525, 101.674443	PPR Pantai Ria	3.097437, 101.671294
PPR Sri Pantai	3.106026, 101.674388	-	-

PPR = Program Perumahan Rakyat.



FIGURE 1. Locations of urban poor communities from Kuala Lumpur in Peninsular Malaysia.

According to SERION ELISA classic, Germany, samples positive for both IgG and IgM antibodies suggest acute infections, although in the present study positive participants did not present any clinical disease symptoms.

Seroprevalences of antibodies to *Leptospira* infections relative to sociodemographic and environmental factors. The sociodemographic profile mainly comprised women (n = 326; 61.3%) and participants of age groups more than 55 years (n = 179, 33.6%). A large proportion was Muslims (n = 480; 90.2%), with more than half unemployed (n = 345; 64.8%). Of the four factors considered (gender, age, religion, and occupation), only age was associated with seropositivity of anti-*Leptospira* IgM ( $\chi_6^2 = 34.145$ , P < 0.01), with seroprevalence being significantly associated with the 13–17 years age group (Table 2).

Environmental health factors considered in the community included the standard of accommodation, sources of drinking water, waste disposal methods, and pet ownership. A significant association was recorded between the seropositivity of anti-*Leptospira* IgG and sources of drinking water ( $\chi_1^2 = 4.087$ ,

P = 0.043), and no risk factors were linked with the seroprevalence of anti-*Leptospira* IgM (Table 3).

Prior knowledge, etiology, and clinical symptoms of leptospirosis. Up to 64.7% of volunteers were knowledgeable in the fundamental aspects of the disease, with 58.6% being aware that leptospirosis was fatal, and 75% appreciated that rats were major carriers of the disease. Those volunteers who lacked knowledge of disease transmission showed significant correlation with seropositivity of anti-*Leptospira* IgM ( $\chi_1^2$  = 5.666, *P* = 0.017) (Table 4).

With reference to etiology, majority of the participants had no contact with the urine of rats (n = 523; 98.3%) and never walked with bare feet (n = 494; 92.9%). A high proportion demonstrated frequent occurrences not only of hand washing (n = 424; 79.7%) but also of hand washing before food consumption (n = 521; 97.9%). A small proportion had neither engaged in aquatic recreational activities recently (n = 24; 4.5%) nor involved in previous flood disasters (n = 55; 10.34%). Overall, none of the issues analyzed were found to

		lgG+		IgM+		IgG+ IgM+	
	Factors	% (95% Cl)	P-value	% (95% Cl)	P-value	% (95% Cl)	P-value
Gender	Male ( <i>n</i> = 206)	9.7 [6.0–14.6]	0.274	2.9 [1.1–6.2]	0.093	0.0 [0.0–0.0]	0.161
	Female ( $n = 326$ )	7.1 [4.5–10.4]		6.1 [3.8–9.3]		0.6 [0.1–2.2]	
Age (years)	< 12 (n = 13)	0.0 [0.0–24.7]	0.737	15.4 [1.9–45.4]	< 0.01*	0.0 [0.0–0.0]	0.317
	13-17 (n = 15)	13.3 [1.7–40.5]		40.0 [16.3-67.7]		6.7 [0.2–31.9]	
	18-24(n = 23)	4.3 [0.1–21.9]		13.0 [2.8–33.6]		0.0 [0.0–0.0]	
	25-34(n=51)	7.8 [2.2–18.9]		0.0 [0.0–7.0]		0.0 [0.0–0.0]	
	35-44 (n = 107)	6.5 [2.7–13.0]		7.5 [3.3–14.2]		0.0 [0.0–0.0]	
	45-54(n = 144)	10.4 [5.9–16.6]		2.1 [0.4–6.0]		0.7 [0.0–3.8]	
	> 55 (n = 179)	7.8 [4.3–12.8]		2.2 [0.6–5.6]		0.0 [0.0–0.0]	
Religion	Islam(n = 480)	8.1 [5.8–10.9]	0.836	4.8 [3.1–7.1]	0.912	0.2 [0.0–1.2]	0.639
0	Buddhist ( $n = 5$ )	20.0 0.5-71.6		0.0 0.0-52.2		0.0 0.0 0.0	
	Hindu ( <i>n</i> = 42)	7.1 [1.5–19.5]		7.1 [1.5–19.5]		2.4 [0.1–12.6]	
	Christian ( $n = 3$ )	0.0 [0.0–70.8]		0.0 [0.0–70.8]		0.0 [0.0–0.0]	
	Others $(n = 2)$	0.0 [0.0-84.2]		0.0 [0.0-84.2]		0.0 [0.0-0.0]	
Occupation	Employed ( $n = 187$ )	7.5 [4.2–12.2]	0.710	3.7 [1.5–7.6]	0.368	0.5 0.0-2.9	0.667
•	Not employed ( $n = 345$ )	8.4 [5.7–11.8]		5.5 3.3-8.5		0.3 0.0-1.6	

TABLE 2 Sociodemographic factors and seroprevalences of IgG+ and IgM+ antibodies to *Leptospira* infections among urban poor communities in Wilayah Persekutuan, Kuala Lumpur

\* Significant at P = 0.05.

be associated with seropositivity of anti-*Leptospira* IgG and IgM (Table 5).

Symptoms of illness among participants, including headaches, jaundice, myalgia, diarrhea, abdominal discomfort, chills, and fevers, appeared to be unrelated to leptospirosis (Table 6).

## DISCUSSION

Leptospirosis has been recognized as a reemerging infectious disease with global importance in urban and rural settings both in tropical and subtropical countries.<sup>18</sup> Leptospirosis remains a major environmental and social disease in the Asia Pacific region with sporadic outbreaks in Southeast Asia, especially in Thailand, India, Malaysia, and Indonesia.<sup>6</sup> The number of human cases worldwide is either underestimated or not well-documented, largely because of inadequate surveillance and similarities in clinical manifestation with other febrile diseases.<sup>19</sup> Malaysia appears to have one of the highest number of disease incidences worldwide,<sup>10</sup> especially in the state of Wilayah Persekutuan, Kuala Lumpur.<sup>11</sup>

The present work represents the first large-scale attempt to determine the seroprevalence of this disease in a targeted community. The discovery of relatively low seroprevalences among urban poor communities in Wilayah Persekutuan, Kuala Lumpur, is an indication of a successful attempt by the Malaysian government to provide basic housing and amenities to these marginalized communities. Nevertheless, there is concern that these communities are at risk of being infected from contaminated water, and although water samples were not examined, the present study has shown a significant association between the seroprevalence of anti-Leptospira IgG with piped water sources. Evidence of pathogenic Leptospira in recreational lakes and sewer effluents has previously been recorded in Kuala Lumpur,20 and many leptospirosis outbreaks have been reported worldwide because of poor sanitation and contamination of urban water supplies.<sup>21-24</sup> In addition, 69 residents from a nurses' hostel in Chennai, South India, were infected with leptospirosis by drinking contaminated water from an underground storage tank.<sup>25</sup> In developing countries, water is treated with chlorine, although recontamination is a major problem often due to the design, construction, and inadequate maintenance of storage facilities and also poor quality control.<sup>26</sup>

Substantial improvements are also needed to improve waste disposal management systems in these residential locations because uncollected garbage provides ideal conditions for rodent reservoir hosts to breed and thrive. A recent

Table 3	
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Environmental health factors and seroprevalences of IgG+ and IgM+ antibodies to *Leptospira* infections among urban poor communities in Wilayah Persekutuan, Kuala Lumpur

		lgG+		lgM+		IgG+ IgM+	
Factors		% (95% Cl)	P-value	% (95% Cl)	P-value	% (95% CI)	P-value
Accommodation	PPR or Flat ( $n = 448$ ) Squatter home ( $n = 84$ )	7.6 [5.3–10.4] 10.7 [5.0–19.4]	0.335	5.1 [3.3–7.6] 3.6 [0.7–10.1]	0.542	0.4 [0.1–1.6] 0.0 [0.0–0.0]	0.407
Drinking water sources	Piped and boiled ( $n = 363$ ) Mineral water ( $n = 169$ )	9.6 [6.8–13.2] 4.7 [2.1–9.1]	0.043*	5.2 [3.2–8.1] 4.1 [1.7–8.3]	0.586	0.6 [0.1–2.0] 0.0 [0.0–0.0]	0.216
Waste disposal near housing area	Yes ( <i>n</i> = 345) No ( <i>n</i> = 187)	8.7 [5.9–12.2] 7.0 [3.8–11.6]	0.481	4.6 [2.7–7.4] 5.3 [2.6–9.6]	0.717	0.3 [0.0–1.6] 0.5 [0.0–2.9]	0.667
Domestic animals at residences	Yes ( <i>n</i> = 182) No ( <i>n</i> = 350)	6.6 [3.5–11.2] 8.9 [6.1–12.3]	0.363	2.7 [0.9–6.3] 6.0 [3.8–9.0]	0.099	0.0 [0.0–0.0] 0.6 [0.1–2.0]	0.195

\* Significant at P = 0.05.

		lgG+		IgM+		lgG+ lgM+	
Prior knowledge		% (95% CI)	P-value	% (95% CI)	P-value	% (95% Cl)	P-value
Basic knowledge	Yes ( <i>n</i> = 344) No ( <i>n</i> = 188)	6.4 [4.1–9.5] 11.2 [7.0–16.6]	0.053	3.2 [1.6–5.6] 8.0 [4.5–12.8]	0.017*	0.3 [0.0–1.6] 0.5 [0.0–2.9]	0.671
Fatal disease	Yes ( <i>n</i> = 312) No ( <i>n</i> = 220)	7.4 [4.7–10.9] 9.1 [5.6–13.7]	0.474	3.5 [1.8–6.2] 6.8 [3.9–11.0]	0.083	0.3 [0.0–1.8] 0.5 [0.0–2.5]	0.805
Transmission by rats	Yes ( <i>n</i> = 399) No ( <i>n</i> = 133)	7.8 [5.3–10.8] 9.0 [4.7–15.2]	0.646	4.3 [2.5–6.7] 6.8 [3.1–12.5]	0.246	0.3 [0.0–1.4] 0.8 [0.0–4.1]	0.447

TABLE 4 Seroprevalences of IgG+ and IgM+ antibodies to leptospirosis relative to prior knowledge of the disease among urban poor communities in Wilayah Persekutuan, Kuala Lumpur

\* Significant at P = 0.05.

study recorded two predominant pathogenic *Leptospira* serovars from two dominant rat species: *Leptospira* borgpetersenii serovar Javanica and *Leptospira* interrogans serovar Bataviae in Kuala Lumpur.<sup>27,28</sup> The presence of pathogenic *Leptospira* thriving in urban rat populations, aided with an abundance of food, poor garbage management, and rapid urbanization, can ultimately result in rodents living in close proximity to human populations, thereby facilitating *Leptospira* transmission.

In the present study, SERION ELISA classic IgG and IgM tests demonstrated a reliable sensitivity of 94.7% and specificity of > 99% compared with the microscopic agglutination test (MAT) (sensitivity of 90%; specificity > 90%) previously shown by Russell et al.<sup>29</sup> and Da Silva et al.<sup>30</sup> This type of serology test, apart from exhibiting a high specificity and sensitivity, typically provides results within 4 hours<sup>31</sup> and also distinguishes acute and previous infections.<sup>32</sup> By contrast, the MAT technique takes several hours to complete<sup>18</sup> and is also unable to differentiate between current, recent, and past infections. Polymerase chain reaction (PCR) methods, on the other hand, offer greater sensitivity and specificity when used in clinical cases because infections can be diagnosed rapidly and completed within the first week of illness.33 But PCR is more costly to run, requiring expensive equipment and skilled laboratory personnel.30

However, positive IgM antibody is a strong indicator of acute *Leptospira* infections.<sup>34</sup> Normally, IgM antibodies can be detected not only 2 days following the onset of symptoms but also up to a further 5 months in infected patients. Yet, not all patients demonstrate IgG production following a *Leptospira* infection. Silva et al.<sup>34</sup> showed that a maximum of

87.5% patients had IgG antibody titers up to 2–3 months postinfection and the remaining patients demonstrated no responses. Therefore, in the present study, participants who were positive for both anti-*Leptospira* IgG and IgM (n = 2) were considered to harbor acute infections.

Young participants, especially within the 13- to 17-year age group, showed a relatively high prevalence of 40% of anti-Leptospira IgM compared with 17.1% in the 20- to 29-year age group in a previous study in Malaysia.<sup>35</sup> In China, a high proportion of students in the younger age group were also diagnosed with leptospirosis likely to be related to the frequency of outdoor recreational activities, including jungle trekking, swimming, kayaking, climbing, and mountain biking.<sup>36,37</sup> On the other hand, in Malaysia from 2004 to 2012, a higher number of leptospirosis cases occurred in the middle-aged 30- to 39-year age group,<sup>35</sup> and these findings are similar to those reported in the Netherlands and Germany, where greater levels of mobility of this age group may increase the risk of exposure.<sup>38,39</sup> Furthermore, increases in the incidences of leptospirosis in more active groups in the community are likely to be associated with recreational activities, such as water sports.13,35,40

In the present investigation, a lack of knowledge on the transmission of leptospirosis was identified as a risk factor, particularly the presence of anti-*Leptospira* IgM and IgG in blood samples examined from a range of participants. Poor awareness of this disease, including inadequate sanitation and hygiene practices, together with the presence of rodent populations contributed to the risk of infection as in other communities.<sup>41–43</sup> Effective cleaning and disinfection, to alleviate

TABLE 5

Seroprevalences of IgG+ and IgM+ antibodies to leptospirosis relative to etiological factors among urban poor communities in Wilayah Persekutuan, Kuala Lumpur

		lgG+		IgM+		IgG+ IgM+	
Etiological factors		% (95% Cl)	P-value	% (95% Cl)	P-value	% (95% Cl)	P-value
Rat's urine contact	Yes (n = 9)	0.0 [0.0–33.6]	0.216	0.0 [0.0–33.6]	0.340	0.0 [0.0–0.0]	0.794
	No (n = 523)	8.2 [6.0–10.9]		5.0 [3.3–7.2]		0.4 [0.0–1.4]	
Walking without shoes	Yes $(n = 38)$	5.3 [0.6–17.7]	0.484	7.9 [1.7–21.4]	0.408	0.0 [0.0–0.0]	0.586
3	No $(n = 494)$	8.3 [0.6–11.1]		4.7 [3.0-6.9]		0.4 [0.1–1.5]	
Involved in flood	Yes $(n = 24)$	16.7 [4.7–37.4]	0.159	0.0 [0.0–14.2]	0.117	0.0 [0.0–0.0]	0.667
	No $(n = 508)$	7.7 [5.5–10.3]		5.1 [3.4–7.4]		0.4 [0.0–1.4]	
Waterfall/river visit	Yes $(n = 55)$	5.5 [1.1–15.1]	0.427	7.3 [2.0–17.6]	0.414	0.0 [0.0–0.0]	0.508
	No $(n = 477)$	8.4 [6.1–11.2]		4.6 [2.9-6.9]		0.4 [0.1–1.5]	
Hand wash	< 3 times ( $n = 20$ )	5.0 0.1-24.9	0.814	5.0 [0.1–24.9]	0.932	0.0 0.0	0.635
	3–5 times (n = 88)	9.1 [4.0–17.1]		5.7 [1.9–12.8]		0.0 0.0	
	> 5 times ( $n = 424$ )	8.0 5.6–11.0		4.7 [2.9–7.2]		0.5 0.1-1.7	
Eating with hand	Yes (n = 521)	8.1 [5.9–10.7]	0.903	5.0 3.3-7.2	0.291	0.4 [0.0–1.4]	0.772
č	No (n = 11)	9.1 [0.2–41.3]		0.0 0.0–28.5]		0.0 [0.0–0.0]	

Recent fever

kutuan, Kuala Lumpur		lgG+		IgM+	•	IgG+ IgM	+
Clinical symptoms		% (95% CI)	P-value	% (95% CI)	P-value	% (95% Cl)	P-value
Headaches	Yes (n = 237)	10.1 [6.6–14.7]	0.122	6.8 [3.9–10.7]	0.075	0.4 [0.0–2.3]	0.877
	No $(n = 295)$	6.4 [3.9–9.9]		3.4 [1.6-6.1]		0.3 [0.0–1.9]	
Jaundice	Yes $(n = 1)$	0.0 [0.0-97.5]	0.681	0.0 [0.0-97.5]	0.751	0.0 [0.0-0.0]	0.931
	No $(n = 531)$	8.1 [5.9–10.8]		4.9 [3.2–7.1]		0.4 [0.0–1.4]	
Myalgia	Yes $(n = 191)$	6.3 3.3-10.7	0.246	4.2 1.8-8.1	0.572	0.0 0.0	0.182
, ,	No $(n = 341)$	9.1 [6.3–12.7]		5.3 3.2-8.2		0.6 0.1-2.1	
Chills	Yes $(n = 42)$	2.4 0.1-12.6	0.104	9.5 [2.7-22.6]	0.191	0.0 0.0	0.566
	No $(n = 490)$	8.6 6.2–11.4		4.5 2.8-6.7		0.4 0.0-1.5	
Diarrhea	Yes (n = 117)	8.5 4.2-15.2	0.836	6.0 [2.4–11.9]	0.543	0.0 0.0	0.318
	No $(n = 415)$	8.0 5.5–11.0		4.6 2.8-7.1]		0.5 0.1-1.7	
Abdominal discomfort	Yes $(n = 140)$	9.3 5.0–15.4	0.548	7.1 [3.5–12.7]	0.165	0.7 0.0-3.9	0.475

0.948

4.1 [2.4-6.5]

4.8 [3.0-7.2]

5.3 [1.5-12.9]

7.7 [5.2–10.7]

7.9 [3.0-16.4]

8.1 [5.8–11.0]

TABLE 6 Seroprevalences of IgG+ and IgM+ antibodies to leptospirosis relative to clinical symptoms among urban poor communities in Wilayah Persekutuan. Kuala Lumpur

infrequent or improper hand washing, are necessary for disease prevention because leptospirosis is primarily spread through direct or indirect contact with contaminated urine. Therefore, those urban poor communities who are at risk of infection in Malaysia must be made aware of the mode of transmission of leptospirosis so that systematic surveillance, prevention, treatment, and control of the disease can be implemented.

No (n = 392)

Yes (n = 76)

No (n = 456)

In conclusion, leptospirosis is among one of the leading zoonotic causes of morbidity worldwide and also accounts for much mortality in vulnerable human populations in both urban and rural settings. Low seroprevalences of leptospirosis infection were found among urban poor communities in Wilayah Persekutuan, Kuala Lumpur, and these findings suggest the importance of introducing public awareness programs on the epidemiology of leptospirosis and particularly the role of rodents as reservoirs of infection. Rodent control management should, therefore, be implemented, especially in urban slum areas where poor sanitation and infrastructure, together with inadequate waste management, can contribute to disease outbreaks.

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0.871

0.3 [0.0-1.4]

0.0 [0.0-0.0]

0.4 [0.1–1.6]

0.432

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