Seroprevalence of Leptospirosis among Town Service Workers in Northeastern State of Malaysia

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Abstract

Introduction: Town service workers are likely to be exposed to the leptospiral infection as a result of their occupational activities. All activities in solid waste management involve risk of the infection at every step in the process from the point where residents handle wastes in the home for collection or recycling, to the point of ultimate disposal.

Aim and Objectives: The purpose of this study was to determine the seroprevalence of leptospirosis among town service workers as there was very little publication on the issue in Malaysia.

Methods: A cross sectional study was conducted among 296 town service workers in northeastern state of Malaysia. The town service workers consisted of four main job categories namely garbage collector, town cleaner, landscaper and lorry driver. Seroprevalence for leptospirosis was determined by microscopic agglutination test (MAT).

Results: All respondents were Malay males with mean age of 42.1 (SD 8.38) years old with mean duration of employment of 15.6 (SD 8.62) years. The overall seroprevalence of leptospirosis was 24.7 % (95 %CI: 19.7, 29.6). The predominant pathogenic serovar was Bataviae (12.3 %). Garbage collectors and town cleaners were shown to have higher prevalence which were 27.4 % (95 % CI: 16.9, 37.9) and 26.0 % (95 % CI: 17.4, 34.5) respectively.

Conclusion: Town service workers are occupational risk group for leptospirosis as indicated by the high seroprevalence.

Keywords: Leptospirosis, seroprevalence, microscopic agglutination test, town service workers

Introduction

Leptospirosis is a disease of global importance. The Caribbean and Latin America, the Indian subcontinent, Southeast Asia, Oceania, and Eastern Europe, are the most significant foci of the disease [1]. The exact number of human cases worldwide is not known precisely. Based on global data collected by International Leptospirosis Society (ILS), there are currently 300,000 to 500,000 severe cases of leptospirosis annually. This figure is probably underestimated and the true extent of leptospirosis remains unknown due to lack of surveillance worldwide [2]. As leptospirosis has a wide variety of clinical manifestations, it is easily confused with many other diseases [2, 3]. Leptospirotic patient may experience confusion, hallucinations, delusion, convulsions and severe bleeding. Cardiac and pulmonary complications are also common. Death may occur towards the end of the second week. Massive bleeding from the alimentary and respiratory tract may also end fatally. Death is virtually limited to icteric cases and in severe cases the mortality may be as high as 15-40%. In those who are not severely ill, recovery takes place in the second week. Fever subsides and the general conditions improve, however, jaundice takes a longer time to clear [4].

Human infections may be acquired through occupational, recreational, or environmental exposures [5]. Although leptospirosis was formerly considered to be primarily related to occupational disease, reports of recreational exposure involving water- sports activities such as swimming, canoeing, or rafting have been increasing [6].

Town service workers are more likely to be exposed to the leptospiral infection as a result of their occupational activities [7-11]. All activities in solid waste management involve risk at every step in the process, from the point where residents handle wastes in the home for collection or recycling, to the point of ultimate disposal. Based on data from various countries, disease and injury risks for solid waste workers are six times more relative risk of infectious disease compared with control baseline populations [7]. Hence, this study was conducted to determine the seroprevalence of leptospirosis among local town service workers.

Methodology

Study design and population

A cross sectional study was conducted in May 2008 in the Municipal Council's town service department of Kota Bharu, the major town in Kelantan, a northeastern state of Malaysia. The Municipal Council of Kota Bharu is essentially an authority which provides public services within its area of jurisdiction comprising an area of 115.64 km^2 and covers a part of the administrative district area of Kota Bharu.

The workers of town service department comprise four job categories namely garbage collector, town cleaner, landscaper and lorry driver. Garbage collector collects garbage from households and bin centres on designated route within the municipality and dumps the garbage from containers onto lorry for further processing and disposal. Town cleaner sweeps roads, drains as well as market area whereas, landscaper performs a range of duties such as transporting and planting new vegetation, mulching, fertilizing, watering, as well as cut and trim grass using either manual hand tools or power-operated equipment. Lorry driver drives lorry for garbage collection and transporting garbage from collection site to the landfill. Sometimes they assist and help garbage collectors to perform their job.

Based on 17.9% seropositive for leptospirosis among town cleansing workers [11] at 95% CI and 20% non response, the estimated sample size required for the study was 272. The sampling frame was the list of 309 workers who have been working in the department for more than six months whereas office and contract workers were excluded from the study.

Blood samples and serologic tests

Informed written consent was obtained from all subjects based on approved study protocol by the Research Ethics Committee (Human), Universiti Sains Malaysia (Reference No: USMKK/PPP/JEPeM[200.4(2.7)]). Consented respondents were interviewed for socio-demographic, job category and duration of employment. Five milliliters of venous blood was obtained and the serum was separated and stored at -20ºC until further processed. All sera samples were tested for the presence of anti leptospiral antibodies using microscopic agglutination test (MAT) following standard methods [4]. The MAT was performed with a panel of live leptospires. *Leptospira* reference cultures were obtained from WHO Collaborating Centre, Queensland Australia. The serovars used in the MAT were *Australis, Autumnalis, Bataviae, Canicola, Celledoni, Grippotyposa, Hardjo, Hebdomadis, Icterohaemorrhagiae, Javanica, Pomona, Pyrogenes, Patoc 1, Sejroe, Tarassovi, Cynopteri, Ballum, and Djasiman.*

Live *Leptospira* cell suspensions representing 18 serovars were added to serially diluted serum specimen in microtitre plates and incubated at 30^{o}C for two hours. Agglutination was examined using dark field microscopy at a magnification of x100. Positive agglutinations was considered when the approximate numbers of free leptospires are <50% of leptospires in the control wells. The titre result was taken as the last dilution that showed <50% of free leptospires compared to control wells. Seropositive leptospirosis respondent was defined as a person who had MAT titre of 1≥100 [12].

Statistical analysis

Data were entered and analyzed using SPSS Version 12.0 (SPSS Inc., Chicago, USA). All continuous variables were described using mean and standard deviations (SD) whereas for categorical variables, frequencies and percentages were presented. Seroprevalence of leptospirosis was described with the 95% confidence interval (CI).

Results

Out of 309 workers eligible for the study, 296 workers were recruited giving a response rate of 95.8%. All respondents were Malay males with mean age of 42.1 (SD 8.38) years old with mean duration of employment of 15.6 (SD 8.62) years. Pertaining to the level of education, 199 (62.7%) of them have only attained lower secondary school. Table 1 shows sociodemographic characteristics of the town service workers.

The overall seroprevalence of leptospirosis was 24.7% (95%CI= 19.729.6). Among the respondents, garbage collectors and town cleaners showed a higher seroprevalence which were 27.4% (95%CI= 16.9- 37.9) and 26.0% (95%CI=17.4- 34.5) respectively (Table 2).

Table 3 shows the distribution of serovars that were determined by positive MAT among 73 seropositive cases. Among them, the most common identified serovar was non-pathogenic *Patoc 1* (39.7%); however the predominant pathogenic serovar identified in this study was *Bataviae* (12.3%). None of the samples were positive for serovars *Celledoni, Grippotyposa, Hardjo, Pyrogenes, Tarassovi, and Ballum.*

Discussion

Findings in this study documented a high prevalence of leptospiral antibody among town service workers. Using similar methods and cutoff value of MAT titre, study among sanitation workers who involved in street sweeping, garbage disposal, drains cleaning, and public latrines cleaning in Madras City reported a similar prevalence (24.8%) [13]. Thus indicates that those workers who involved in sanitation activities are at risk for leptospiral infection.

In our study, a higher prevalence were noted among garbage collector (27.4%) followed by town cleaner (23.8%), landscaper (13.8%) and lorry driver (17.9%). These were generally comparable with the study done by Chan *et al.* (1987) in Singapore which found that the highest seropositive rates was among garbage collectors (22.2%), followed by market cleaners (22.2%) and street cleansing workers (17.6%) [14]. The higher seroprevalence among garbage collectors probably related to their daily occupational activities. Garbage collectors had a longer exposure and close contact with garbage that might be contaminated with *Leptospira* infected urine compared to other job categories.

In this study, non-pathogenic *L.biflexa* serovar Patoc 1 was the most common serovar identified (39.7%). Similar finding was noted in Turkey among 72 leptospirosis cases [15]. *L.biflexa* serovar Patoc 1 was used as a saprophytic strain which cross reacts with human antibodies generated by a number of pathogenic serovars, in a case of rare or perhaps to a strain that is currently unknown in the area concerned [16]. This may suggest the circulation of other pathogenic serogroups, which may not have been included in the MAT antigens set used for this study.

The predominant pathogenic serovars identified in this study was Bataviae (12.3%), followed by Javanica (11.0%), and Djasiman (8.2%). Study by Swapna *et al.,* (2006) among sewage workers in North Kerala, India demonstrated serovar Betaviae and Djasiman as predominant serovars which was similar compared to this study [17]. Formerly, it was considered that distinct clinical features were associated with specific serogroups [18], but this view was questioned and more intense study over the past 30 years have refuted this hypothesis [5]. However, many other studies on leptospirosis also include study on animal reservoirs [19, 20]. This allows us to compare the serovar frequency between humans and reservoirs, as it tends to be associated and probable transmission route can be drawn. In relation to our study, we could not speculate on reservoirs and sources of infection as animal surveys among rodents and domestics animals have not been done in these area.

During the course of data collection, all respondents were asymptomatic. Asymptomatic infection of leptospirosis is common and has been reported in many studies [14, 19, 21, 22]. The importance of subclinical or asymptomatic infection is not well understood, and efforts to determine its significance were limited [23]. However it is generally believed that serovar-

specific antibodies are protective and the person is immune to re-infection with the same serovar as long as the concentration (titre) of specific antibodies is high enough [16, 21]. In view of that, asymptomatic infections may play a role in population immunity (herd immunity) against leptospirosis. Therefore, seroprevalence study in this high risk group may reflect exposure but not necessarily overt disease. However, in a small proportion of infected persons, severe complications may set in [4]. In such cases, there would be clinical manifestations of multiple organ involvement and the case-fatality rates in different parts of the world have been reported to range from <5% to 30% (25). Mortality remains significant, related both to delays in diagnosis due to lack of infrastructure and adequate clinical suspicion [24]. There has also been the recognition of an emerging, potentially epidemic disease representing a significant public health challenge [25].

Our study had several limitations. Firstly, as the study was performed in a single population, among town service workers in Kota Bharu, our findings may not be generalized to other population. Generalization of our result is limited to Malay, male town service workers at Kota Bharu. Secondly, pertaining to the serological test that has been used, although the microscopic agglutination test (MAT) is the reference method for epidemiological serosurveys of leptospirosis, it has several drawbacks. Misclassification of persons as seronegative or seropositive may be a potential bias [23]. For instance, we might get false negativity in the early course of disease and when the causative *Leptospira* serovar is not included in the panel of typing organisms which do not represent those present in the community as well as the cross-reaction between the antibodies [12]. The cross-reactive of antibodies may be associated with syphilis, human immunodeficiency virus (HIV), legionellosis, malaria and dengue [26]. Unfortunately, we were not able to test our subjects for those diseases due to time, and money constraints. Hence negative test results observed in immunoassays do not necessarily rule out the disease among a small proportion of patients [4]. Furthermore, the cut-off value of MAT usually depends on the baseline in the community in a particular geographical area [4] and varies from lab to lab [12]. Currently, there is no consensus regarding the standard cut-off titre in view of local scenario. The cut-off titre 1≥100 is generally accepted [12] and is often employed in other seroprevalence studies [17, 27, 28]. However, Shivakumar and Krishnakumar recommended using a titer of 1≥50 as cutoff value for serosurveys in the asymptomatic high risk group [29]. In view of that, the seroprevalence in our study might be underestimated.

In conclusion, high seropositivity rates indicate that town service workers are the occupational risk group for leptospiral infection. The workers are constantly exposed to the contaminated environment which suitable for the survival of leptospires and these working conditions and work habits put them under risk of leptospiral infection. Further study on animal (reservoirs) is recommended to provide important information on predominant serovars which allows us to see the pattern and interaction of human and *Leptospira* host.

Conflict of interest statement

We declare that we have no conflict of interest.

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Table 1: Sociodemographic characteristics of town service workers (n= 296)

Variable	Frequency (%)	Mean (SD)
Age (year)		42.1 (8.38)
Marital status		
Married	262 (88.5)	
Single	32 (10.8)	
Widower	2 (0.7)	
No of children		3.7 (2.53)
Income (RM)		1252 (317)
Level of education		
Primary school or lower	64 (21.6)	
Lower secondary school	135 (45.6)	
Upper secondary school	95 (32.1)	
Form Six or higher	2 (0.7)	
Job category		
Town cleaner	104 (35.1)	
Landscaper	80 (27.0)	
Garbage collector	73 (24.7)	
Lorry driver	39 (13.2)	

Table 2: Seroprevalence of leptospirosis using MAT among 296 town service workers according to job categories

Job category	No. of workers	MAT 1≥100	
		Frequency (%)	95% CI
Garbage collector	73	20 (27.4)	16.9 - 37.9
Landscaper	80	19 (23.8)	14.2 - 33.3
Town cleaner	104	27 (26.0)	17.4 - 34.5
Lorry driver	39	7 (17.9)	5.3 - 30.6
TOTAL	**296**	**73 (24.7)**	**19.7 - 29.6**

Pearson Chi-square test [p-value = 0.713 (df = 3)]

Table 3: Serovars distribution determined by positive MAT (titre 1≥100) among 73 seropositive town service workers

Serovars tested	Frequency	Percentage (%)
Patoc 1	29	39.7
Bataviae	9	12.3
Javanica	8	11.0
Djasiman	6	8.2
Canicola	5	6.8
Cynopteri	4	5.5
Australis	3	4.1
Hebdomadis	3	4.1
Icterohaemorrhagiae	3	4.1
Autumnalis	1	1.4
Sejroe	1	1.4
Pomona	1	1.4