ISSN: 2252-8806, DOI: 10.11591/ijphs.v14i4.25797

Work practices linked to seropositive leptospirosis among cattle farmers in Northeastern Malaysia

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Article Info

Article history:

Received Aug 25, 2024 Revised Sep 3, 2025 Accepted Nov 3, 2025

Keywords:

Cattle farmers Leptospirosis Occupational health Risk factors Seropositivity

ABSTRACT

Leptospirosis is a re-emerging zoonotic disease with significant global health implications, particularly in tropical and subtropical regions. In Malaysia, the warm, humid climate and frequent exposure to livestock and contaminated environments increase the risk of infection, especially among agricultural workers. Cattle farmers regularly handle animals and work in unsanitary conditions, which puts them at heightened risk. This study aimed to determine the prevalence of leptospirosis seropositivity and identify risk factors associated with contracting leptospirosis among cattle farmers in Northeastern Malaysia. A cross-sectional study was conducted involving 120 cattle farmers in Northeastern Malaysia. Data were collected through an interviewer-guided questionnaire, and serological testing was performed using the microscopic agglutination test with a seropositive cut-off titre of ≥1:100. The prevalence of leptospirosis seropositivity was found to be 72.5%. Significant risk factors included working with a wounded hand (Adj. OR: 7.26; 95% CI: 2.61-20.18; p<0.001), working with a wounded leg (Adj. OR: 8.52; 95% CI: 1.98-36.66; p=0.004), not wearing rubber gloves (Adj. OR: 3.96; 95% CI: 1.13-13.91; p=0.032), and not showering immediately after work (Adj. OR: 6.04; 95% CI:1.69-21.62; p=0.006). The high seroprevalence of leptospirosis among cattle farmers indicates a significant occupational risk. Future prevention programs should prioritize promoting safe work practices to mitigate this risk.

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INTRODUCTION

Leptospirosis is a re-emerging zoonotic disease with global relevance, particularly widespread in tropical and subtropical areas. It poses a significant threat to public health, affecting millions of individuals annually, with case fatality rates ranging from 5% to 30% [1]. However, the exact number of cases has yet to be precisely determined due to a lack of worldwide surveillance [2]. For example, during outbreaks, incidence rates can surpass 100 cases per 100,000 individuals in high-risk populations [3]. Current estimates suggest around 1.03 million cases (95% CI: 434,000-1,750,000) and approximately 58,900 deaths (95% CI: 23,800-95,900) from leptospirosis occur globally each year [4]. However, these figures are believed to be

significantly underestimated due to factors such as limited public awareness, non-specific clinical symptoms, insufficient diagnostic tools, and underdeveloped surveillance systems [5], [6].

In Malaysia, leptospirosis is endemic, with numerous outbreaks and a notable mortality rate reported over the past decade [7], [8]. The country's year-round tropical climate, coupled with seasonal monsoons and heavy rainfall, creates favorable conditions for the survival of the Leptospira bacteria. The bacteria can persist for weeks to months in warm, moist soil and on water surfaces, contributing to a high incidence of the disease [9], [10]. Carrier animals disseminate the bacteria into the environment, where it can infect humans either through direct contact with infected urine or indirectly through contaminated soil or water. Human-to-human transmission is exceedingly rare as humans are incidental hosts and do not typically contribute to onward transmission [11], [12]. The disease is often associated with occupational exposure, particularly affecting those in agricultural professions. Workers in this sector, especially those in close contact with livestock, are recognized as being at heightened risk of contracting leptospirosis [13], [14]. A study highlighted that close interaction between humans and livestock, particularly in traditional farming systems, contributes to significant health risks. Cattle farmers, in particular, have been identified as a critical group for targeted preventive interventions [15].

The livestock sector plays a pivotal role in Malaysia's economy, fulfilling domestic needs for meat, milk, and dairy products while employing rural communities. Recent national statistics indicate an increasing livestock population, particularly in rural areas of northeastern Malaysia, underscoring the importance of addressing zoonotic risks in this sector [16]. Despite this, there remains a gap in understanding the specific factors contributing to leptospirosis transmission among cattle farmers, a group with distinct environmental and occupational exposures. This study aimed to determine the prevalence of leptospirosis seropositivity and to identify the risk factors for contracting leptospirosis among cattle farmers in Northeastern Malaysia.

2. METHOD

This cross-sectional study was carried out among cattle farmers across six districts in Northeastern Malaysia. The districts were chosen through simple random sampling from a pool of ten districts, ensuring an unbiased and representative selection process. A list of cattle farmers from these districts was obtained from the Department of Veterinary Services. Stratified random sampling was then used to allocate the number of farmers from each district, ensuring proportional representation across different regions. This method ensured the sample captured the geographic diversity of cattle farming practices in the area, effectively reducing selection bias.

The sample size was calculated using the estimated 37.5% seroprevalence of leptospirosis among animal handlers, reported in previous studies [17]. Considering a potential non-response rate of 30%, the total sample size was increased to 120 to ensure sufficient power for statistical analysis. Eligible participants were those registered with the Department of Veterinary Services and had worked as cattle farmers for at least six months, which allowed us to focus on experienced farmers potentially exposed to risk factors over a longer period. Farmers not registered or working less than six months were excluded to reduce variability in exposure levels.

Participants provided written informed consent before the study and were interviewed using a validated, structured questionnaire in Malay. The questionnaire was designed to gather data on sociodemographic information (age, gender, education level) and work practices (use of personal protective equipment, exposure to animals and water sources). Venous blood samples were collected from each participant and sent to the Institute for Medical Research (IMR) for analysis using a microscopic agglutination test (MAT) to detect leptospiral antibodies. The MAT was conducted with a panel of live leptospires, with reference cultures obtained from the Biomedical Research Royal Tropical Institute in Amsterdam and IMR. Agglutination was defined as $\leq 50\%$ free leptospires (i.e., $\geq 50\%$ agglutination) compared to the control well. A titre of $\geq 1:100$ was used as the leptospirosis seropositivity cut-off, indicating past leptospira exposure [8], [18].

Data were entered and analyzed using IBM SPSS Version 22 software, with confidentiality maintained throughout the analysis. Continuous variables were described using mean and standard deviation (SD), while categorical variables were summarized using frequencies and percentages. The seroprevalence of leptospirosis was reported with a 95% confidence interval (CI). Univariable analysis was done using simple logistic regression in order to analyze work practice-related risk factors for leptospirosis seropositivity. All variables that showed p-value <0.25 were considered important and selected to be further tested using multiple logistic regression. The preliminary final model was obtained after variable selection using Forward Likelihood Ratio (Forward LR) selection and Backward Likelihood Ratio (Backward LR) elimination methods. No significant interactions or multicollinearity were observed in the final model. The Hosmer–Lemeshow test (Chi-square = 3.676, p = 0.751) indicated that the model fit well. The model's overall

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classification accuracy was 80.0%, which correctly predicted 80.0% of the cases. Additionally, the area under the ROC curve was 0.817 (95% CI: 0.731, 0.903), reflecting strong discriminatory ability.

3. RESULTS AND DISCUSSION

This study examined the prevalence and risk factors of leptospirosis among cattle farmers in Northeastern Malaysia, revealing several significant findings. Table 1 presents the sociodemographic characteristics of the respondents. All participants were Malays, with a mean age of 50.5 years (SD = 14.94). The majority were married (78.3%), and the average family size was 5.2 members (SD = 2.37). Most respondents reported a monthly income of less than RM 1000 (60.8%), and 10.0% had no formal education.

Table 1. Sociodemographic characteristics of the respondents (n = 120)

Variables	Frequency (%)	Mean (SD)			
Age (years)		50.5 (14.94)			
Gender					
Male	104 (86.7)				
Female	16 (13.3)				
Marital status					
Married	94 (78.3)				
Single/widower	26 (21.7)				
Family Members		5.2 (2.37)			
Income					
<rm1000< td=""><td>73 (60.8)</td><td></td></rm1000<>	73 (60.8)				
RM1000 to RM2000	32 (26.7)				
>RM2000	15 (12.5)				
Education level					
No formal education	12 (10.0)				
Primary school	31 (25.8)				
Secondary school	77 (64.2)				

The seroprevalence of leptospirosis among cattle farmers in this study was remarkably high at 72.5% (95% CI: 63.5, 80.1). This prevalence significantly surpasses that reported in other occupational groups. For instance, previous studies have documented seroprevalence rates of 32.6% among oil palm plantation workers, 25.5% among hospital staff, 23.2% among rubber estate workers, and 17.9% among town cleaning laborers [19]. Additionally, a study conducted in Kelantan, Malaysia, identified agriculture workers as having the highest prevalence of leptospirosis within various occupational categories [14]. Comparatively, healthy paddy planters in northeastern Malaysia exhibited a seroprevalence of 24.2% using the same serological test (MAT) [20], and military recruits in Peru after jungle training showed a 28.0% seroprevalence [21]. A recent study by Philip and Ahmed found that agricultural workers, particularly those in livestock farming, consistently show higher seroprevalence rates for leptospirosis compared to other occupational groups across various geographical regions [6]. Meanwhile, a study by Sohm et al. [22] provides a comprehensive European perspective on leptospirosis in cattle, highlighting the ongoing significance of this zoonotic disease across different geographical regions. These findings indicate that cattle farmers are at two to three times higher risk of leptospirosis compared to other high-risk occupational groups. This elevated risk is likely due to the constant presence of cattle and other livestock, which maintain environmental reservoirs of Leptospira through sustained micturition.

3.1. Risk factors associated with seropositivity

Table 2 presents the association between work practice characteristics and leptospirosis seropositivity among cattle farmers. The multiple logistic regression model identified four independent predictors of infection risk, underscoring the occupational hazards faced by this group. Farmers who did not wear rubber gloves during work were nearly four times more likely to be seropositive compared to those who consistently used gloves (AOR = 3.96; 95% CI: 1.13–13.91; p = 0.032). Likewise, sustaining hand wounds while working markedly increased the odds of seropositivity by more than sevenfold (AOR = 7.26; 95% CI: 2.61–20.18; p <0.001), while leg wounds conferred an even higher risk (AOR = 8.52; 95% CI: 1.98–36.66; p = 0.004). In addition, failure to shower immediately after work was associated with a sixfold higher likelihood of seropositivity (AOR = 6.04; 95% CI: 1.69–21.62; p = 0.006). These findings highlight the critical role of personal protective equipment and post-work hygiene in interrupting exposure pathways. The elevated risks linked to open wounds suggest that Leptospira organisms can readily penetrate compromised skin, whereas the protective effect of gloves and immediate bathing emphasizes the importance of creating physical and behavioral barriers to infection in high-exposure environments.

3.2. Implications of high seroprevalence and identified risk factors

The significantly higher seroprevalence of leptospirosis among cattle farmers compared to other occupational groups and the general population underscores the heightened vulnerability of this group to Leptospira infection. In the present study, the seroprevalence of 72.5% among cattle farmers was markedly higher than the 8.4% prevalence reported in a Malaysian hospital-based survey of febrile patients [14], reflecting the intense environmental exposure faced by this occupational group. This elevated risk is consistent with evidence from India, where agricultural and livestock workers have demonstrated significantly higher seroprevalence rates than the general population [23]. The convergence of these findings across different geographic settings highlights the universal nature of occupational risk, suggesting that close and repeated contact with livestock and contaminated environments is a key driver of infection. These results reinforce the need for targeted prevention strategies, including consistent use of personal protective equipment, routine health surveillance, and workplace hygiene interventions, to reduce the burden of leptospirosis among high-risk farming communities.

A notable finding is the high prevalence of working with wounds among cattle farmers. Over half of the respondents reported having worked with hand wounds, and a quarter with leg wounds. The nature of cattle farming, involving the use of sharp equipment and exposure to rough environments, predisposes workers to such injuries. These wounds serve as entry points for Leptospira, as supported by studies from Germany, Nigeria, India, and Thailand [24]-[27]. The increased odds of seropositivity associated with wounds on the hands (AOR = 7.26) and legs (AOR = 8.52) highlight the critical role of physical injuries in leptospiral transmission. Recent research by Bierque et al. on Leptospira survival in different environmental conditions suggests that the bacteria can persist longer on moist skin, potentially explaining the increased risk associated with open wounds [10]. Similarly, Bradley and Lockaby's review on leptospirosis and the environment further underscores the complex interplay between environmental conditions and disease transmission, emphasizing the need for targeted interventions in high-risk settings like cattle farms [2].

Furthermore, the protective effect of wearing rubber gloves (AOR = 3.96) emphasizes the importance of personal protective equipment (PPE) in mitigating infection risk. This aligns with outbreak reports, such as the one following a major flood in Australia, where none of the confirmed leptospirosis cases had worn protective gloves [28]. Additionally, Selim *et al.* [29], who studied bovine Leptospirosis in Egypt, emphasized the importance of personal protective equipment in reducing infection risk among livestock handlers. Similarly, the significant protective effect of showering immediately after work (AOR = 6.04) suggests that personal hygiene practices are crucial in reducing environmental contamination and preventing bacterial entry through wounds. A study on Leptospirosis risk factors and management challenges in developing countries highlights immediate and thorough washing after potential exposure as a key preventive measure [30].

Interestingly, handwashing with soap after work was not a significant protective factor in this study. This contrasts with findings from palm oil plantation workers in Malaysia, where handwashing was protective [23]. The discrepancy may be attributed to the broader exposure of cattle farmers to environmental risk factors beyond the hands, such as through leg wounds, making handwashing insufficient on its own. Comprehensive hygiene practices, including full-body washing, appear to be more effective in high-exposure settings like cattle farming.

3.3. Significance and future directions

The high seroprevalence of leptospirosis among cattle farmers highlights an urgent public health concern. The implications of these findings extend beyond Malaysia, as evidenced by recent data from other regions. For example, the global relevance of these findings is further supported by Barnabé *et al.* [31], who provided new insights into the diagnosis and epidemiology of bovine leptospirosis in Brazil's Caatinga biome, highlighting the need for improved surveillance and control measures in diverse agricultural settings, In addition, recent surveillance data from the European Centre for Disease Prevention and Control 2022 show an increase in leptospirosis cases across Europe, with occupational exposure being a significant risk factor [32]. It suggests ongoing exposure to Leptospira in this occupational group, which may represent repeated exposure rather than overt disease, as individuals can be reinfected with different serovars despite existing antibodies [33], [34]. This finding underscores the necessity for targeted interventions, including the consistent use of PPE, regular health screenings, and education on personal hygiene practices tailored to the unique risks of cattle farming.

Future research should explore the relationship between wound severity and leptospirosis seropositivity to better understand the mechanisms of transmission in high-exposure farming environments. Investigating whether factors such as wound size, depth, duration of exposure, and the presence of secondary skin infections influence the likelihood of Leptospira penetration would help clarify the biological pathways of infection and inform targeted protective measures. In addition, well-designed intervention studies are needed to evaluate the effectiveness of comprehensive PPE protocols and enhanced hygiene practices including routine use of gloves, waterproof boots, and full-body washing on reducing infection rates among

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cattle farmers. Such evidence would provide a robust scientific basis for developing occupational health guidelines, strengthening policy implementation, and optimizing resource allocation for disease prevention in this vulnerable workforce.

Table 2. Association of work practice characteristics of the respondents with seropositive leptospirosis using

simple and multiple logistic regression analyses (n = 120)

Seropositive			Cr. OR ^a	Adj. OR ^b	P-value*	
Variables	(n = 87)	n = 33 (95% CI)		(95% CI)		
	n(%)	n(%)				
Wear rubber boots						
Yes	65 (72.2)	25 (27.8)	Ref			
No	22 (73.3)	8 (26.7)	1.01 (0.42, 2.69)			
Wear rubber gloves	, í	`				
Yes	8 (47.1)	9 (52.9)	Ref	Ref	0.032	
No	79 (76.7)	24 (23.3)	3.70 (1.29, 10.65)	3.96 (1.13, 13.91)		
Wear long pants						
No	8 (23.5)	26 (76.5)	Ref			
Yes	79 (91.9)	7 (8.1)	0.90 (0.34, 2.42)			
Wear a long-sleeved shirt						
No	25 (75.8)	8 (24.2)	Ref			
Yes	62 (71.3)	25 (28.7)	0.79 (0.32, 1.99)			
Working with a wounded hand	, í	, ,				
No	26 (54.2)	22 (45.8)	Ref	Ref	< 0.001	
Yes	61 (84.7)	11 (15.3)	4.69 (1.99, 11.06)	7.26 (2.61, 20.18)		
Working with a wounded leg						
No	59 (66.3)	30 (33.7)	Ref	Ref	0.004	
Yes	28 (90.3)	3 (9.7)	4.75 (1.33, 6.89)	8.52 (1.98, 36.66)		
Showering immediately after work	` /	` /	. , ,			
Yes	8 (44.4)	10 (55.6)	Ref	Ref	0.006	
No	79 (77.5)	23 (22.5)	4.29 (1.52, 12.14)	6.04 (1.69, 21.62)		
Washing hands with soap	` ′	. /		, , ,		
Yes	57 (64.8)	31 (35.2)	Ref			
No	30 (93.8)	2 (6.2)	4.63 (1.02, 21.04)			
Smoking	` ′	. ,				
No	26 (68.4)	12 (31.6)	Ref			
Yes	61 (74.4)	21 (25.6)	0.51 (0.22, 1.16)			

a: Simple logistic regression, b: Multiple logistic regression, Hosmer and Lemeshow Test, p-value = 0.751, No multicollinearity and no interaction were found, Classification Table overall percentage = 80.0%, Area under ROC curve = 81.7% (95% CI: 73.1, 90.3)

4. CONCLUSION

The high seroprevalence of leptospirosis among cattle farmers suggests that they are at considerable risk of contracting the disease. The nature of their work, which involves frequent contact with soil and water likely contaminated by the urine of infected animals, significantly increases this risk. Future prevention programs should focus on implementing safe work practices among cattle farmers, including mandatory use of personal protective equipment, regular wound care, and comprehensive hygiene protocols. Additionally, targeted education programs and routine health screenings for this high-risk group are crucial for reducing the incidence of leptospirosis.

While this study provides valuable insights, it is important to acknowledge its limitations. The cross-sectional design limits causal inferences, and the focus on a specific region may affect generalizability. Future longitudinal studies across diverse geographical areas could provide more comprehensive data on the dynamics of leptospirosis transmission among cattle farmers.

ACKNOWLEDGMENTS

The authors would like to express their deepest gratitude to Assoc. Prof Dr. Mohd. Mokhtar Arshad from the Faculty of Veterinary Medicine, University Malaysia Kelantan, and Dr. Suratan Kamarudin from Kelantan Veterinary Services Department for their contribution to this research. This study was funded by the Research University Grant (1001/PPSP/8012288). Finally, thanks to all of the study respondents who provided us with insightful responses.

FUNDING INFORMATION

This study was funded by the Research University Grant (1001/PPSP/8012288) from Universiti Sains Malaysia.

Int J Public Health Sci ISSN: 2252-8806 1811

AUTHOR CONTRIBUTIONS STATEMENT

This journal uses the Contributor Roles Taxonomy (CRediT) to recognize individual author contributions, reduce authorship disputes, and facilitate collaboration.

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C : Conceptualization Vi: Visualization I : Investigation Su: Supervision M : Methodology R: Resources So: Software D: Data Curation P : Project administration Va: Validation O: Writing - Original Draft Fu: Funding acquisition

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

INFORMED CONSENT

Written informed consent was obtained from all participants prior to their inclusion in the study. All personal identifiers were removed to maintain confidentiality and privacy.

ETHICAL APPROVAL

Ethical approval was granted by the Research and Ethics Committee (Human), Health Campus Universiti Sains Malaysia (USM/JEPeM/15050164).

DATA AVAILABILITY

The datasets generated and/or analyzed during the current study are not publicly available due to privacy and confidentiality restrictions but are available from the corresponding author on reasonable request.

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