Seroepidemiological Profile and Risk Factor Analysis of Bovine Leptospirosis in Buffaloes from Bhola District, Bangladesh

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Abstract: Leptospirosis is a cosmopolitan zoonotic disease distributed in both humans and animals. The present study was conducted in the selected Bathan region of Bhola district, Bangladesh, from July 2022 to June 2023. A total of 160 buffaloes were investigated by rapid diagnostic test (*RGenomix Bovine Leptospira Ab* Rapid Detection Test Kit, India). The overall prevalence of bovine leptospirosis in buffaloes was 5%. The highest prevalence of the disease was recorded in individuals aged>5 years at 5.71%. The present study also showed that the highest prevalence of bovine leptospirosis was recorded in buffaloes with the highest parity >4(10.52%), and the low BCS group <1-2(9.37%). According to management factors, the infection of leptospirosis in buffaloes was highest in poor management conditions (12.30%) and large-sized Bathan of buffaloes (10.25%). In addition, the prevalence of bovine leptospirosis was comparatively higher in dairy buffaloes (5.83%) than in dry conditions. The infection rate in pregnant buffaloes is 15% and in non-pregnant buffaloes is 3.57%. The findings of the study might be helpful to assess the economic importance and preventive measures of the disease in the study area.

Keywords: Seroepidemiology, risk factors, leptospirosis, buffaloes

Introduction

Leptospirosis is a zoonosis of worldwide distribution. The disease is caused by infection with pathogenic Gram-negative bacteria of the genus *Leptospira*, which is classified into pathogenic and nonpathogenic species with more than 250 pathogenic *serovars* (Chadsuthi et al., 2018). The incidence is highest during rainy seasons in tropical regions and during summer and autumn in temperate regions (Suwancharoen et al., 2013). Human cases typically involve agricultural workers, rice field workers, vacationers, and plumbers (Palla et al., 2025). Cases of *leptospirosis* in children and adults exposed to diseased livestock or contaminated water have been documented (Kurtoglu et al., 2003, Al Abaidani et al., 2016). *Leptospira* spp. infections are acquired either through direct exposure to the urine or tissues of infected animals or through indirect contact with contaminated environments, particularly moist areas. Domestic animals act as important reservoirs and help maintain the bacteria in nature. *Leptospira* can be found in various domestic mammals, including rodents, cattle, sheep, dogs, cats, and other livestock (Sunaryo and Priyanto, 2022). Human interaction with these animals increases the risk of disease transmission.

Rodents are known to be the main reservoir of leptospirosis (Nova et al., 2020). This waterborne illness impacts hundreds of millions of people globally, many of whom are unable to avoid exposure, whether they live in rural or urban environments in developed and developing countries alike. Although waterborne leptospirosis is not transmitted by the fecal–oral route as are cholera, bacterial dysentery, and other diarrheal diseases (Bharti et al., 2003). The transmission of *leptospirosis* occurs within an ecological context of humans coming into contact with water contaminated by urine from a wide variety of both wild and domesticated animals. Humans and animals are most infected by *Leptospira* through contact with the urine or birth products (e.g., foetus, placenta, amniotic fluids) of an infected animal or contact with *Leptospira*-contaminated surface waters, mud, or soil (Vinetz et al., 2005). The presence of the organism has been demonstrated serologically in a wide variety of livestock that includes cattle, sheep, and goats (Adler and de la Peña Moctezuma, 2010).

A wide spectrum of clinical signs is associated with leptospirosis in different animal species, reflecting its extensive zoonotic host range. Comparable clinical presentations occur in cattle and buffalo and are linked to substantial economic losses. Acute disease in livestock may lead to reproductive failure, abortion, stillbirth, lowered milk yield, infertility, and occasionally death; however, subclinical infections constitute the majority. Mastitis, along with altered milk flow and quality, may arise, during which *Leptospira* can be excreted in milk. Chronic leptospirosis and carrier status with colonization of the renal tubules by *Leptospira* for extended periods of time and intermittent shedding of the organism in urine commonly occur in livestock (Suwancharoen et al., 2013).

Leptospirosis is still a neglected tropical disease in Bangladesh. There is a need to improve awareness and understanding of leptospirosis among individuals at risk of exposure, as well as among local healthcare personnel. Demonstrating the presence of environmental exposure risks is expected to help enhance this awareness (Krijger et al., 2019). There is limited research available on bovine leptospirosis in livestock populations of coastal areas. Based on these considerations, the present study was conducted to evaluate the seroepidemiology and risk factors of bovine leptospirosis in buffaloes in the specified region. In addition, the research aimed to assess the prevalence of leptospirosis among various reservoir hosts, particularly domestic animals such as rodents and livestock.

Materials and Methods

Study area and duration

This study was performed in Bhola district, Bangladesh, where managemental factors were visible to determine the seroepidemiology and risk factors of bovine leptospirosis in buffaloes from July /2022 to June /2023.

Diagnosis of the disease

The preliminary diagnosis of bovine leptospirosis in buffaloes was made using the owner's complaints, disease history, general physical evaluation, clinical signs, and results obtained from a rapid field screening test designed for bovine leptospirosis.

History taking

A detailed case history was obtained by interviewing the owner, farmer, or caretaker to gather information related to disease onset, symptoms, and management. For hygienic management, water and waste management, rodent control, fencing swamp or wet area, and isolate affected individuals from healthy cattle.

General physical examination

The animals were assessed visually to record overall health status, behavior, occurrence of abortion or stillbirth, birth of weak calves, and any other systemic abnormalities (renal failure, anemia, anorexia, depression etc.).

Clinical examination

Clinical evaluation of the buffaloes was carried out using history, presenting complaints, clinical observations, and standard leptospirosis diagnostic techniques.

Serological test An immune chromatographic rapid diagnostic test was performed on the collected serum samples using the Genomix Bovine *Leptospira* Rapid Detection Test Kit (GENOMIX CARL PVT. LTD.), following the manufacturer's protocol.

Statistical analysis

Data obtained from the field and laboratory investigations were entered into Microsoft Excel 2007. Univariable associations between explanatory variables and sero-positivity were evaluated using the chi-

square test. It implies explanatory or independent variables (age, parity, BCS, lactation status, physiological status, hygienic management and Bathan size) influence the disease occurrence.

Results

This study provides information about the prevalence and risk factors of bovine leptospirosis in buffaloes in Bhola district. In addition, understand the progression and effect of the disease by abortion, stillbirth, decreased milk production, infertility, and death.

Prevalence of bovine leptospirosis in buffaloes

A total of 160 suspected buffalo samples were examined to determine the prevalence of bovine leptospirosis. Among these, 8 samples tested positive, resulting in an overall prevalence of **5.0%** in the study area (Figure 1).

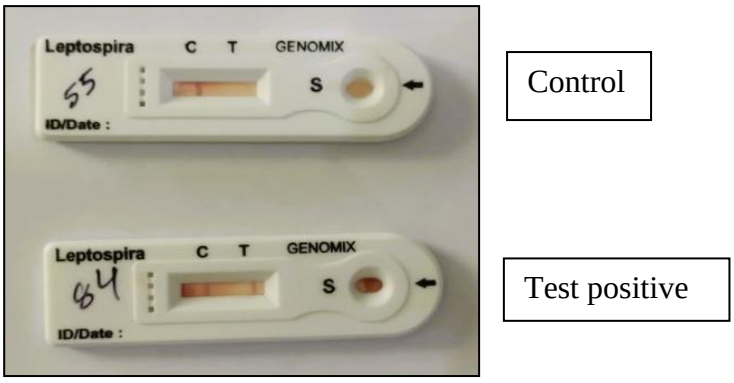

Figure 1. Upper lane showing control without test band, on the other hand, lower lane showing both control and test positive band (Rapid Test)

Demographic determinants of bovine leptospirosis in buffaloes

Table 1. Seroprevalence of bovine leptospirosis in buffaloes according to demographic factors.

Demographic determinants	Categories	Number tested	Positive sample	Prevalence (%)	Chi-square	p-value
Age	<5 years	55	2	3.64	0.036	0.8486
	>5 years	105	6	5.71		
Parity	1	28	1	3.57	1.442	0.6957
	2	49	2	4.08		
	3	64	3	4.68		
	>4	19	2	10.52		
BCS	>3-4	19	0	0.00	5.041	0.0804
	>2-3	85	2	2.35		
	>1-2	64	6	9.37		
Lactation status	Milking buffaloes	120	7	5.83	0.175	0.6753
	Dry buffaloes	40	1	2.50		
Physiological status	Non-pregnant	140	5	3.57	2.707	0.0999
	Pregnant	20	3	15		

In Table 2, the highest prevalence of leptospirosis was recorded in the >5 years of age group (5.71%) compared with the <5 years of age group (3.64%). In case of parity, the highest prevalence was recorded in >4 among the other parity groups. According to body condition score, the highest prevalence of leptospirosis in buffaloes was recorded in the BCS <1-2 group at 9.37% followed by BCS >2-3 (2.35%) and BCS >-3-4 (0%). In case of lactation status, comparatively, milking buffalo cows (5.83%) were more susceptible than dry cows (2.50%)**.** According to physiological status, pregnant buffaloes (15%) were more susceptible than non-pregnant buffaloes (3.57%).

Bovine leptospirosis in buffaloes according to management factors

Table 2. Seroprevalence of bovine leptospirosis in buffaloes according to management factors

Management factor	Categories	Number tested	Positive sample	Percentage (%)	Chi-square	p-value
Hygienic management of the farm	Good	40	1	4.00	1.734	0.4203
	Medium	55	2	5.81		
	Poor	65	5	12.30		
Bathan size	<30-50	82	3	5.85	0.19	0.6633
	>50	78	5	10.25		

In Table 3. According to hygienic management of the Bathan, the Bathan with poor management (12.30%) was highly susceptible to bovine leptospirosis in buffaloes, followed by medium (5.81%) and good (4.00%) management conditions. In case of Bathan size, animal numbers >50(10.25%) were comparatively more infected than animal numbers <30-50 animal numbers (5.85%) were recorded in the study area.

Discussion

Prevalence of bovine leptospirosis in buffaloes

The overall prevalence of bovine leptospirosis in buffaloes was 5%. (Table 1 and Figure 1). Patel *et al*. reported the seroprevalence of leptospirosis in buffaloes as 15.69% (Patel et al., 2016) . Another recent study revealed that 91.5% of buffaloes tested positive for anti-leptospira antibodies in the northern part of Brazil (Barbosa et al., 2023), where the seroprevalence rate was higher than in the present study. In addition, the seroprevalence of leptospirosis in dairy cattle was 47.27% which was higher than the reported study (Parvez et al., 2015). Researchers from other countries also reported varied prevalence of leptospirosis in cattle, such as 20.3 % in Sri Lanka (Gamage et al., 2011), 19.1% in Iran (Tabatabaeizadeh et al., 2011), and 27.4% in New Zealand (Subharat et al., 2011). The variation might be due to different geographical locations, management practices, disease resistance, and levels of natural immunity (Sykes et al., 2022)

Bovine leptospirosis in buffaloes according to demographic variables.

According to age-wise distribution (Table 2), the higher prevalence of leptospirosis was recorded in the >5 years of age group (5.71%) than the <5 years age group (3.64%), as shown in Table 2.

The seroprevalence of leptospirosis tended to increase with the age of animals (Dos Santos et al., 2012) which supports the present study. In addition, Parvez *et al*. reported the highest prevalence of bovine leptospirosis in dairy cattle of more than 7 years of age, and inclined to the present study (Parvez et al., 2015). The animals with older age might have decreased immunity, which increases their susceptibility to infection.

In parity-wise distribution (Table 3), the highest prevalence was recorded in >4 (10.52%). The present study was in support of Parvez *et al*. (Parvez et al., 2015). Another study showed that the parity number has no effect on the prevalence of leptospirosis in dairy cattle (Desa et al., 2021). Gompo *et al*. reported that the cows with parity <2 were comparatively more susceptible than parity >2, which reverses the present study (Gompo et al., 2020).
In addition, the highest prevalence of leptospirosis in buffaloes was recorded in the BCS <1-2 group at 9.37% followed by BCS >2-3 (2.35%) and BCS >-3-4 (0%) (Table 4). A research group reported that there was no significant effect of body condition score on the seroprevalence of bovine leptospirosis in cattle (Roberts et al., 2010).
According to lactation status, comparatively milking buffalo cows (5.83%) were more susceptible than dry cows (2.50%) shown in Table 5. It has been reported that the milking cows (53.5%) were comparatively more infected with leptospirosis than dry cows (30.4%) (Parvez et al., 2015), which supports the present study. Gompo *et al*. also found that lactating cows were more susceptible to leptospirosis than nonlactating cows (Gompo et al., 2020).
The present study also reported that (Table 6) pregnant buffaloes (15%) were more susceptible than non-pregnant buffaloes (3.57%). According to the physiological status of dairy cattle, it has been reported that fresh cows (78.5%) were more infected with bovine leptospirosis than cyclic (56.5%) and pregnant cows (40%) (Parvez et al., 2015), which is the reverse of the reported data. However, another study found that there was no significant effect of the physiological status of dairy cows on the seroprevalence of leptospirosis in dairy cows (Gompo et al., 2020).

Bovine leptospirosis in buffaloes according to management factor

According to hygienic management of the Bathan, the Bathan with poor management (12.30%) was highly susceptible to bovine leptospirosis in buffaloes, followed by medium (5.81%) and good (4.00%) management conditions (Table 7). A study reported that the cows reared in a poor hygienic environment were highly susceptible to leptospirosis (Desa et al., 2021), which inclined the present study. They also found that the farms where rodents are readily available were most susceptible to the disease. Another study showed that animal feed contaminated with soil was more infected with bovine leptospirosis than in noncontaminated conditions (Gompo et al., 2020).
According to Bathan size, animal numbers >50(10.25%) were comparatively more infected than <30-50 animal numbers (5.85%) (Table 7). It has been reported that large and medium-sized herd sizes of cattle were more infected with leptospirosis than small herd sizes (Desa et al., 2021). Gompo *et al* also reported that the animals in large herds were more susceptible to leptospirosis than small herds (Gompo et al., 2020).

Conclusion

Leptospirosis is a zoonotic disease that endangers both animal and human health. The demographic findings showed that that bovine leptospirosis was more prevalent in older animals, animals with higher parity, poor body condition, pregnant animals, and milking buffaloes. In addition, poor hygienic management practices and large herd sizes were identified as major factors increasing the susceptibility to leptospirosis in buffaloes. Further detailed molecular epidemiological research is needed to target female buffaloes for future control and surveillance programs in the study area.

Recommendations

Enhanced hygienic management and prioritized health monitoring is needed in older and high-parity buffaloes. Regular seroprevalence and immunoprophylaxis shouldbe maintained properly to check the infection.

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Authors Contributions

Md. Selim Ahmed conceptualized and designed the study. Mizanur Rahman drafted the manuscript. All authors have reviewed and approved the final version of the manuscript.

Conflict of interest

None to declare

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