

Original paper

Prevalence and factors influencing gastrointestinal parasitic infections in sheep in Bangladesh

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ABSTRACT. Gastrointestinal (GI) parasites are major contributors to decrease productivity in livestock over the world. A cross-sectional study was conducted in different areas of Bangladesh to determine the prevalence of GI parasitic infections and their association with the biotic and abiotic factors in sheep. A total of 572 faecal samples were collected from the selected areas of Bangladesh and microscopic examination was performed for the identification of parasites using flotation and sedimentation technique. Out of 572, 441 animals were found infected with one or more species of GI parasites with an overall prevalence of 77.1%. Nine types of parasites from four different classes were detected namely Strongyles (42.1%), *Strongyloides* sp. (27.1%) and *Trichuris* sp. (1.0%), *Moniezia* sp. (2.4%), *Paramphistomum cervi* (32.5%), *Fasciola gigantica* (6.1%) and *Schistosoma* sp. (3.5%), coccidia (16.6%) and *Balantidium coli* (7.9%). Nematodes infections (56.8%) were significantly highest among trematodes (37.9%), protozoa (24.4%) and cestode (2.4%). In the present study, all the biotic factors including sex, age, physiological condition of female and body condition score (BCS) of animals were insignificantly ($p>0.05$) associated with the prevalence of GI parasitic infection in sheep but among the abiotic factors, muddy housing of animals, rainy season, having no knowledge about GI parasites and illiteracy of farmers were significantly ($p<0.05$) associated with the GI parasitic infections. This epidemiological investigation will assist to build a suitable control program against GI parasites in sheep and thus, help to prevent production loss and increase livelihood of small holder farmers.

Key words: factors, prevalence, GI parasites, sheep, Bangladesh

Introduction

Sheep are important part of livestock and reared in different parts of the world due to its excellent adaptation capability in different climatic condition [1]. This sector also provides employment to many millions of people all over the world, and especially in the underdeveloped and developing countries. In 2015, according to the United Nations' Food and Agriculture Organization, global sheep population estimated at approximately 1.17 billion and almost 37% sheep are reared in Asia [2]. Farming of sheep is becoming more popular among young entrepreneurs in Bangladesh as it earns more profit with a small investment. Now a days, 3.15 million sheep are available in Bangladesh [3] and play a key role in global food systems as the main source of animal protein such as milk, meat [1]. It also plays

an important role in poverty alleviation of resource-poor and privilege deprived people.

But production performance of animal is greatly hampered and do not reach the optimal level due to gastrointestinal parasitism, one of the most common infection in livestock [4]. In sheep, these can vary from subclinical weight loss to clinical symptoms such as anemia, diarrhoea and severe protein loss and ultimately death [5]. The severity of disease is mainly influenced by factors such as the parasite fauna present, worm burden in the gastrointestinal tract, general health condition and immunological status of the host, and environmental factors, such as climate and pasture type, stress, stocking density, management and/or diet [6–10]. The climatic condition of Bangladesh is suitable for the growth and development of parasites. As a result, different types of parasites such as nematodes, cestodes,

trematodes and protozoa are common in sheep under both rural and farm conditions in Bangladesh [11,12].

These parasitic infection have negative effects on productivity such as delayed growth, decreased live weight gain or weight loss, reduced reproductive performance such as fertility and condemnation of meat [13,14]. Regarding these effects, they cause a major impact on the economy of the country [15,16]. In our previous communication, a detailed epidemiological investigation was performed for gastrointestinal nematodes in goats [17]. Although sheep and goats both are small ruminants having similar type of management system, breeding and nutritional status of these species but their feeding nature is different, for example, goats are browser (take food from the top) and sheep are bottom feeder, therefore, it is quite logical that sheep are more likely to be infected. For this, a comprehensive epidemiological knowledge of gastrointestinal parasites in sheep in Bangladesh is essential to control the parasitic diseases in this area. By considering this point, the present study was aimed to investigate the prevalence of gastrointestinal parasites and their associated risk factors in sheep.

Materials and Methods

Study area and design

A cross sectional study was conducted to estimate the overall prevalence of GI parasites in sheep in Bangladesh from July, 2016 to June, 2017. Geographically, Bangladesh lies in the north eastern part of South Asia between latitudes 20°34'N and 26°38'N and between longitudes 88°01'E and 92°41'E. The sample collecting areas were selected according to the availability of sheep rearing including Madhupur (Tangail district), Godagari (Rajshahi district), Badarganj (Rangpur district), Mymensingh Sadar (Mymensingh district), Shailkopa (Jhenaidah district), Char Fasson (Bhola district) and Rangamati Sadar (Rangamati district) (Fig. 1). A subtropical monsoon climate is present in Bangladesh and three seasons can be distinguished; namely the cool-dry winter (November to February), the hot dry summer (March to June) and the hot-wet rainy season (July–October) [18].

Sample size

Approximately the total sheep population size in the study areas were 18,888. The sample size for population survey was calculated by using the

Statcalc function of EpiInfo v.7.2.3.1 (CDC, Atlanta, USA). Having the expected proportion of 67.9% [11,19] and 5% margin of error, the estimated sample size was 562 at confidence level of 99%. A non-response rate of 10% was considered, and thus a total of 624 sheep were selected by using disproportionate stratified random sampling from seven study areas. However, the final selection was based on the household owner's willingness to cooperate. Fifty-two owners declined, and therefore, 572 sheep were included in the study.

Data collection

Data relevant to biotic and abiotic factors such as sex, age, body condition score (BCS), farming system, flock size, farming nature, housing, knowledge about GI parasites and education level of farmers were recorded in a structured questionnaire. Age of the animal was categorized into three groups: 1–6 months, >6–18 months and >18 months following eruption chart of teeth and also by interviewing farmers [20]. The physiological condition of females was grouped into pregnant, lactating and non-pregnant. Body condition score of animals were categorized as poor body conditioned animals ($BCS \leq 2$) and good body conditioned animals ($BCS > 2$) following the parameters described previously [21]. Briefly, in $BCS \leq 2$, spinous processes and transverse processes are sharp and prominent or slightly rounded and it is possible to pass figure under the end of the transverse processes with little or no pressure. In $BCS > 2$, spinous processes or transverse processes can be or cannot be detected without pressure and loin eye muscle is full with fat. The management system was recorded by visiting farmers' houses. The farming system of animals was categorized as backyard and semi-intensive; flock size was grouped as ≤ 5 , 6–20 and > 20 ; farming nature of animal management was categorized into mixed and single farming, and housing was categorized as muddy and concrete/ slatted. The farmers' information such as knowledge about GI parasites and educational level of farmer was recorded by interviewing with the owners of the animals (Supplementary file 1).

Collection of samples and microscopic examination

A total of 572 faecal samples were collected directly from the rectum of each animal using hand gloves in an airtight glass vial with 10% formalin.

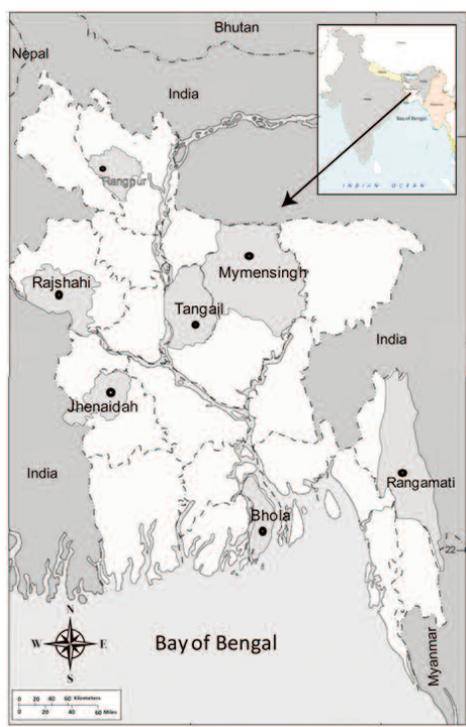


Figure 1. Map of Bangladesh. Circle indicates sample collecting areas.

Samples were immediately shifted into the laboratory, Department of Parasitology, Bangladesh Agricultural University, Mymensingh-2202. Samples were processed for microscopic examination using flotation technique for nematode/cestode eggs and protozoan cyst/oocyst and simple sedimentation technique for trematode eggs. Briefly, in flotation technique, about 2g of faeces was weighed and taken in a test tube. Then, flotation fluid (saturated salt solution, specific gravity-1.200) was added, mixed thoroughly, strained through sieve (50 meshes) and poured on another test tube. The test tube was filled with saturated salt solution up to the brim, placed a cover slip over the meniscus and allowed to stand for 10 minutes to float eggs/cysts/oocysts. After which, the cover slip was removed and placed on a glass slide and examined under compound microscope (LABOMED, los Angeles, CA, USA) using 10 \times objective. In simple sedimentation technique, about 2g of faeces was weighed and taken into a beaker with 10 ml tap water. A homogenous mixture was made, strained through a sieve and poured in a centrifuge tube. Sample was centrifuged at 1500 rpm for 20 minutes, and then the supernatant was discarded carefully. After loosening the faecal pellet, a few drop of sediment diluting with a drop of water was taken on a glass slide, covered with cover slip and examined

under compound microscope using 10 \times objective [22]. Identification of helminth eggs and protozoan cyst/oocyst were done following keys and description given by Soulsby [23] and Thienpont et al. [24].

Statistical analyses

The data were analyzed into computer program, SPSS version 20.0. Chi-square test was employed to estimate the strength and statistical significance ($p < 0.05$) of associations between predictor variables and GI parasitic infection.

Ethical statement

No animals were unethically injured/killed during the research period. The study was approved by Animal welfare and ethical committee of Bangladesh Agricultural University (06/AWEC/2017).

Results

Overall prevalence of GI parasites in sheep

An epidemiological investigation on GI parasites was carried out in 572 animals in different areas of Bangladesh. The present study revealed that a spectrum of GI parasites was available in naturally infected sheep in these study areas and out of 572 animals, 441 (77.1%) animals found to be infected with one or more species of parasites. Nine parasites

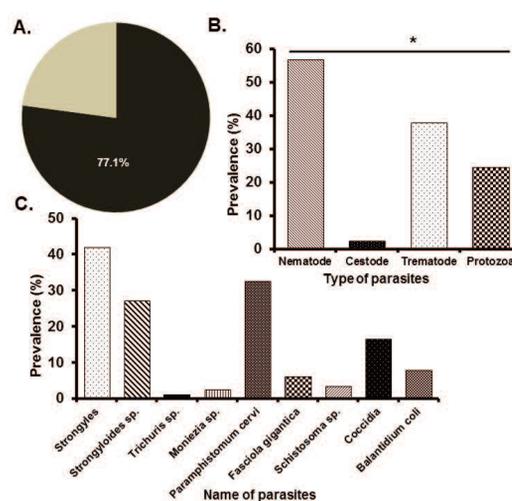


Figure 2. Prevalence of parasites in sheep in Bangladesh. Faecal samples from sheep (n=572) were collected and preserved in 10% formalin, and examined using flotation and sedimentation techniques. A. Overall prevalence of gastrointestinal parasites, B. Prevalence of nematode, cestode, trematode and protozoa (* $p < 0.05$), C. Prevalence of different parasites.

Table 1. Univariate analysis of biotic factors for gastrointestinal parasites in sheep

Factors	Level	Infected/Examined	Prevalence (%)	OR
			(95% CI)	(95% CI)
Sex	Male	141/180	78.3 ^a (71.7-83.7)	1.108 (0.73-1.69)
	Female	300/392	76.5 ^a (72.0-80.4)	
Age	1-6 months	26/31	83.9 ^a (67.3-92.9)	1.70 (0.63-4.61)
	7-18 months	196/250	78.4 ^a (72.8-83.0)	1.19 (0.79-1.78)
	>18 months	219/291	75.3 ^a (69.9-79.8)	
Physiological condition of female	Pregnant	41/48	85.4 ^a (72.8-92.7)	2.09 (0.83-5.07)
	Lactating	171/225	76.0 ^a (70.0-81.1)	1.11 (0.67-1.86)
	Non pregnant	88/119	73.9 ^a (65.4-81.0)	
Nutritional status	Poor	208/262	79.4 ^a (74.0- 83.8)	1.27 (0.85-1.88)
	Good	233/310	75.2 ^a (70.0-79.6)	

Explanations: Values with different letters within a column in each variable differ significantly ($p < 0.05$); OR, Odds Ratio; CI, Confidence Interval

were identified. Of them, three were nematodes such as *Strongyles* (42.1%), *Strongyloides* sp. (27.1%) and *Trichuris* sp. (1.0%), one cestode such as *Moniezia* sp. (2.4%); three trematodes such as *Paramphistomum cervi* (32.5%), *Fasciola gigantica* (6.1%) and *Schistosoma* sp. (3.5%), and two protozoa such as coccidia (16.6%) and *Balantidium coli* (7.9%). Only one cestode, *Moniezia* sp. was identified in our experiment. Significantly highest infections were caused by nematodes (56.8%) followed by trematodes (37.9%), protozoa (24.4%) and cestode (2.4%) (Fig. 2).

According to spatial distribution of GI parasitic infection, highest infection was recorded in Godagari, Rajshahi (92.3%) and lowest infection in Madhupur, Tangail (61.4%). The details of spatial distribution of parasites were shown in Fig. 3.

Influence of biotic factors on the prevalence of GI parasitic infection in sheep

In our study, we observed that males (78.3%) were more prone to GI parasitic infection than females (76.5%). However, this variation was statistically insignificant ($p > 0.05$). Epidemiological information according to age revealed that prevalence of GI parasitic infection were decreased by increasing the age of animals and it was

statistically insignificant ($p < 0.05$). The prevalence of parasitic infections was higher in pregnant (85.4%) and lactating (76.0%) than non pregnant (73.9%) animals. According to body condition score, poor body condition of animals were 1.27 times more susceptible to GI parasitic infection than good body condition of animals (Table 1).

Influence of abiotic factors on the prevalence of GI parasitic infection in sheep

In the present study, according to abiotic factors, farming system, flock size and farming nature of animal management were insignificantly ($p > 0.05$) associated with the GI parasitic infections. However, muddy housing of animals, rainy season, having no knowledge about GI parasites and illiteracy of farmers were significantly ($p < 0.05$) associated with the GI parasitic infections. The risk of GI parasitic infections was more than two-fold higher in animals owned or managed by muddy housing system (83.5%) compared to concrete/slatted housing system (69.3%). Rainy season (86.1%) was considered the most vulnerable season than that of summer (76.3%) and winter (68.9%) season. Also, the risk of GI parasitic infections were 1.74 times and 1.66 times more in those farmers who had no knowledge about GI parasites (79.0%)

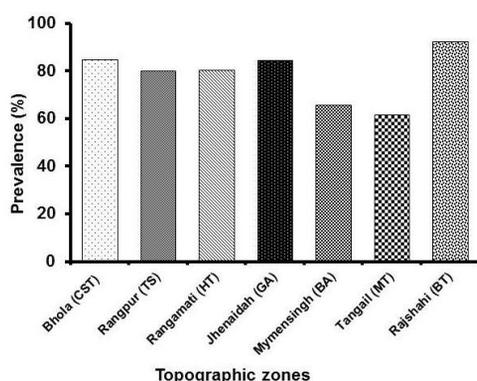


Figure 3. Prevalence of gastrointestinal parasitic infections in sheep in different topographic zones in Bangladesh. Faecal samples from sheep (n=572) from seven topographic zones such as Coastal Saline Tract (CST, n=107), Tista Silt (TS, n=101), Hill Tract (HT, n=77), Gangatic Alluvium (GA, n=64), Brahmaputra Alluvium (BA, n=96), Madhupur Tract (MT, n=88), Barind Tract (BT, n=39) were collected and preserved in 10% formalin, and examined using floatation and sedimentation techniques. Prevalences were estimated.

and no educational background (79.5%) than those farmers having knowledge about GI parasites (68.3%) and educational background (69.9%), respectively (Table 2).

Discussion

The epidemiological investigation on GI parasitic infection in sheep have been conducted in different parts of the world [6–8,25] due to their impact on animal health and production [23]. They had a significant impact on the economic balance of a country [15] in terms of subclinical infection, loss of meat and wool, treatment and control cost and mortality of animals [26].

Distribution of GI parasitic infection is influenced by the climatic condition of an area. The development and survival of free living stages of parasites depend on temperature, humidity, light intensity and rainfall [27]. Parasitic infection especially GI parasites in ruminants are most common in temperate and humid climate [28]. In our study, we found that 77.1% sheep were infected

Table 2. Univariate analysis of abiotic factors for gastrointestinal parasites in sheep

Factors	Level	Infected/examined	Prevalence (%) (95% CI)	OR (95% CI)
Farming system	Backyard	337/432	78.0 ^a (73.8-81.6)	1.22 (0.78-1.91)
	Semi-intensive	104/140	74.3 ^a (66.4-80.8)	
Flock size	≤5	330/421	78.4 ^a (74.2-82.0)	1.29 (0.82-2.04)
	6-20	16/22	72.7 ^a (51.8-86.8)	
	>20	95/129	73.6 ^a (65.4-80.4)	
Farming nature	Mixed	45/53	84.9 ^a (72.9-92.1)	1.74 (0.80-3.80)
	Single	396/519	76.3 ^a (72.4-79.7)	
Housing	Muddy	263/315	83.5 ^a (78.9-87.1)	2.24 (1.50-3.34)
	Concrete/ slatted	178/257	69.3 ^b (63.3-74.5)	
Season	Rainy	179/208	86.1 ^a (80.7-90.1)	2.79 (1.71-4.54)
	Summer	116/152	76.3 ^b (68.9-82.3)	
	Winter	146/212	68.9 ^b (62.3-74.7)	
Knowledge about GI parasites	No	372/471	79.0 ^a (75.0-82.4)	1.74 (1.08-2.79)
	Yes	69/101	68.3 ^b (58.7-76.5)	
Education level	Illiterate	341/429	79.5 ^a (75.4-83.0)	1.66 (1.08-2.55)
	Literate	100/143	69.9 ^b (61.9-76.8)	

Explanations: see Table 1

with different types of GI parasites.

The subtropical monsoon climate of Bangladesh is suitable for the development and survival of free living stage, maintained mainly in pasture. Taylor et al. [13] reported that optimum environmental temperature (18–26°C) and moisture (80–100%) regulated embryonation, hatching and survivability of developmental stages of parasites. Although, the development process of parasites is arrested below temperature 10°C.

The most common genera of GI parasites in sheep in this experiment were nematode including *Strongyles*, *Trichuris* and *Strongyloides*; cestode including *Moniezia*; trematodes including *Fasciola*, *Paramphistomum* and *Schistosoma*, and protozoa including coccidia and *Balantidium*. A wide range of GI parasites that belong to these genera have been reported in different regions of the world with similar climatic condition [6–8].

In the present study, we observed that males were more likely to be infected with GI parasitic infection than female. Similar findings were also observed by Islam et al. [29], Zvinorova et al. [30] and Tariq et al. [25] who attributed this to the genetic predisposition and differential susceptibility owing to hormonal control.

Lambs (young) are known to be more susceptible than adults and there is a tendency to decrease percentage of worm infection in animals with the increase of age [31]. The young animals are naïve in any infection including parasitic infection and adults are protective due to acquired immunity through frequent exposure [32]. More parasitic infection in male could be attributed to genetic predisposition and differential susceptibility owing to hormonal control i. e. testosterone [33].

The physiological stress condition in female during late pregnancy and lactation decrease the resistance of the hosts to parasites and consequently resulting higher prevalence of parasites [34]. Body condition score is linked to the immune response of host. Malnourished animals are more susceptible to any infections, including GI parasitic infection and the fecundity of parasites is increased in malnourished animals [35–37]

According to abiotic factors, muddy housing, rainy season, having no knowledge about GI parasites and illiteracy of farmers were significantly associated with GI parasitic infection.

In Bangladesh, animals were mostly reared by backyard system. Traditionally, different age groups and different farm animals such as cattle, sheep and

goats share same pasture land as it is generally the main source of food for these animals. Thus, contaminated pasture plays an important role for transmitting parasitic infection in grazing animal. Housing system is the potential risk that significantly associated with GI parasites in sheep, supporting the result of Hassan [38]. In muddy housing system, mud, water, urine and faecal materials made an unhygienic condition with optimum temperature and moisture that favor parasitic development especially hatching, development, survival and transmission of parasites [39].

The small holder marginal farmers in Bangladesh are mostly from low educational background and have no proper knowledge about GI parasites. They usually rear animals by extensive system and do not maintain proper farming system or deworming schedules. Moreover, majority of the farmers do not show any interest in receiving veterinary service for improving their management systems due to lack of adequate financial solvency. Thus, the parasitic infection is more in animals reared by farmers with illiteracy and having no proper knowledge about GI parasites [40].

In this study, strongyles had the highest prevalence and that of nematodes, trematodes and cestodes varied significantly. Among the biotic and abiotic factors, muddy housing, rainy season, having no knowledge about GI parasites and illiteracy of farmers were significantly associated with GI parasitic infection in sheep. Therefore, housing, and farmer's knowledge, attitude and practice should be improved for controlling the parasites and thus, to reduce production loss and improve livelihood of small holder farmers.

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Supplementary file 1

Data sheet for faecal sample

- SL No.: _____ Date: _____
 Name of owner: _____ Mobile no.: _____
 Address: _____
 Faecal sample collection: _____
1. Area:
 - a. Coastal Saline Tract (CST-Bhola)
 - b. Tista Silt (TS-Rangpur)
 - c. Hill Tract (Rangamati)
 - d. Gangatic Alluvium (GA-Jhenaidah)
 - e. Brahmaputra Alluvium (BA-Mymensingh)
 - f. Madhupur Tract (Tangail)
 - g. Barind Tract (BT-Rajshahi)
 2. Sex: Male/ Female
 3. Age: 1-6 month/>6-18 month/≥18 months
 4. Physiological condition of female:
 - Pregnant/Non pregnant/Lactating
 5. Body condition: Poor/Good
 6. Farming system: Backyard/Semi-intensive
 7. Flock size: ≤5/6-20/≥20
 8. Farming nature: Single (only sheep)/Mixed (Sheep/goats/cattle)
 9. Housing: Concrete or Slatted/Muddy
 10. Knowledge about the harmful effects of GI parasites? Yes/No
 11. Education level of farmer: Illiterate/Literate
 12. Season: Summer/winter/rainy
 13. Findings: _____
- Signature _____