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Seroprevalence of brucellosis in small ruminants in selected area of Bangladash

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Abstract

A seroprevalence study of small ruminant brucellosis was conducted in sheep and goat rearing selected areas of Mymensingh district and Dhaka district, Bangladesh, from March, 2005 to May, 2006. Sera from 62 sheep and 300 goats were tested by rose bengal plate test (RBPT), plate agglutination test (PAT), tube agglutination test (TAT) and mercaptoethanol test (MET). Out of the 62 sera tested 3.25% (n = 2) were positive to RBT, PAT and TAT and 4.84% (n = 3) were positive MET. In case of 300 goats, 1.67% (n = 5) were positive to RBT and PAT, 2% (n = 6) were positive to TAT and 2.33% (n = 7) were positive to MET. This investigation is the first of its type to be performed in small ruminants in Bangladesh. Higher prevalence rate (8.0 %) was found in BAU nutrition farm in case of sheep and 10 % in Bangladesh Agricultural University (BAU) Veterinary Clinic in case of goat while lower prevalence (0.0 %) was recorded in Pharmacology project and BAU adjacent villages in case of sheep and (0.0 %) in Dhamrai upazila in case of goats respectively. *Brucella* antibodies were more prevalent in sheep (8.84 %) than in goat (2.33 %).

Key Words: Brucellosis, Small ruminants, Villages (Boyra and Char Nilaykha), Ishwargonj Upazila and Dhamrai upazila.

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Introduction

Bangladesh hosts large number of small ruminants that are raised usually under free range system or in adjunct to crop production. The ruminants especially small (sheep and goat) ruminants in Bangladesh are mainly utilized for meat purposes, although goat milk is used some extent for human consumption. The small ruminants are not only important for meat and milk but also important for good quality leathers and source of income to farmers. Among the Asiatic countries Bangladesh has got the second highest population of goats which accounted for 34.47 million [1]. The goat rank second in terms of meat, milk and skin production representing about 28.0, 23.0 and 28.0 per cent among the total contribution of livestock, in Bangladesh [1]. There are about 33.55 million goats and 1.16 million sheep in Bangladesh [2]. The sheep and goats can significantly play an important role in the economic well being of the resource-poor farmer. Moreover, the sheep and goats enterprise is becoming more popular due to socio-economic condition and their ability to survive on poor quality pastures and forage that is unsuitable for other species of ruminants. Besides, goats require relatively small investment and can therefore, be a source of cash income for small-scale farmers. The disease constrains make hindrance for the development of goat industry and there is a lot of report for abortion but there is no report whether the abortion is due to brucellosis. In spite of the presence of huge small ruminant population, Bangladesh fails to optimally utilize this resource as the sector is suffering from lower productivity. Among many factors that limit the economic return from small ruminant production diseases stand in the front line. One of such diseases that hamper the productivity of small ruminants is brucellosis.

Brucellosis is a wide spread disease of livestock and human beings resulting in reproductive inefficiency and abortion. Small ruminant brucellosis is mostly caused by *Brucella meltensis* [3]. *B ovis* is also an important cause of orchitis and epididymitis in sheep but it is not recognized as a cause of natural infection in goats. Persistent infection is a common feature of the disease with frequent shedding of the bacterium in reproductive and mammary secretions. Brucellosis is an important zoonosis threatening the public health in many countries of the world [4]. The risk of brucellosis is presumed to be high in nomadic pastoral societies where close and frequent contact between man and animals is unavoidable part of the ecology.

Brucellosis has been reported in small ruminants from different parts of the world. Prevalence rates of 1.7% in sheep and 1.5% in goats in Sudan, 6.01% in sheep and 2.8% in goats in Kenya, 5.29% in goats and 7.2% in sheep in Somalia [5], 3.8% in goats and 1.4% in sheep in Eritrea [6], 4% in goats and 1% in sheep in eastern Sudan, 6.6% in sheep and 4.75% in goats in Nigeria [7] have been

recorded. From 255 sheep and 289 goats slaughtered at an abattoir of New Dehli India, brucellosis was diagnosed in 9.02 %, 4.31%, 27.45% and 10.95% sheep and 1.73%, 1.38%, 7.27% and 18.34% goats using rose bengal plate test (RBPT), standard tube agglutination test (STAT), complement fixation test (CFT) and dot-ELISA, respectively [8]. However, in Bangladesh only one study in the reported the presence of brucellosis in small ruminants with prevalence of 0% in sheep and 1% in goats [9] and prevalence of caprine brucellosis were determined 23.64% at Modhupur in Tangail district, 31.82% at Bhaluka in Mymensingh district, 34.0% in Manikganj district and 16.66% in BAU campus and adjacent villages by PAT [10]. Despite the presence of large population of small ruminants in Bangladesh no work has been carried out on brucellosis.

The objective of this study was, therefore, to determine the prevalence of small ruminant brucellosis in selected areas of Bangladesh.

Materials and Methods

The study was conducted for a period of 15 months from March, 2005 to May, 2006 in the Department of Medicine, Faculty of Veterinary Science, Bangladesh Agricultural University (BAU), Mymensingh.

1. Sampling strategies

Blood samples were collected from sheep and goat of different areas of Bangladesh. In Mymensingh, BAU Nutrition Farm, goats attended at Veterinary Clinic, BAU and the sheep and goat population living around the BAU campus were included in this study. Besides, samples were collected sera from sheep and goat of Boyra, Char Nilakhia, Vangnamari villages of Ishwargonj upazilla, Mymensingh and some villages of Dhamrai upazila, Dhaka. The sexually matured female sheep and goat populations were randomly selected for this study. All of the study animals were indigenous breeds. No *Brucella* vaccine has been used in both of the study areas. The study recorded some e.g. age, pregnancy and non-pregnancy etc information in individual animal. All the blood samples were processed for sera preparation and were tested with the rose bengal test (RBT) and plate agglutination test (PAT) as screening tests and tube agglutination test (TAT) and mercaptoethanol test (MET) test as confirmatory tests.

2. Experimental animals

A total of 362 animals of foresaid location (300 goats and 62 sheep) were selected randomly for blood collection.

3. Blood and sera samples collection

About 3-5㎖ of blood was collected from jugular vein of each goat and sheep with the help of sterile disposable syringe and needle. Later on, the sera were separated in test tube from each labeled syringe and the test tube was marked with same number by permanent marker. Then the sera were centrifuged at 2,000 rpm for 10 minutes. After centri-

fugation a clear sera were found and then the sera were transferred to the sterilized labeled eppendorf tube. The vial was stored in ice chamber at −20 ℃ for future use.

4. Serological study

RBT, PAT and TAT were used for the diagnosis of brucellosis. Animals found positive by RBT and PAT and the negative reactors both were further confirmed by the TAT and MET.

5. RBT

The preparation of diagnostic antigen and procedure were conducted according to the procedure of Baek et al [11]. The prepared antigen was standardized according to the procedure of OIE, 2000. Briefly inactivated S11193 whole cells were washed with 0.5% phenol saline (0.85%) and suspended in 0.5% phenol saline (0.85%) 1 g to 22.5mℓ. Thirty five mℓ of this suspension was added to 1mℓ of 1% Rose Bengal (Sigma Co, USA) and stirred for 2 hrs at room temperature. The stained cells were uniformly resuspended 1 g of cells to 7mℓ of diluent (21.1g of sodium hydroxide dissolved in 353mℓ of phenol saline, followed by 95mℓ of lactic acid, and adjusted to 1,056mℓ with phenol saline). After filtration through cotton wool, it was adjusted to a packed cell volume of 8 %.

Sera samples and the antigen were brought to room temperature. Then thirty microliter of serum was mixed with the equal volume of antigen on a clear glass plate circled approximately 2cm in diameter with manicure. The mixture was rocked gently for 4 min at room temperature, and then observed. Any sign of agglutination was considered positive [12].

6. PAT

The preparation of diagnostic antigen and procedure were conducted according to the procedure of Ryu et al [13]. The prepared antigen was standardized according to the procedure of OIE [14]. Briefly, inactivated S1119−3 whole cells were washed with 0.5 % phenol saline (0.85 %) and suspended at 11.0 % instead of 4.5 of TAT. Crystal violet brilliant green staining solution was prepared by dissolving 2g brilliant green and 1g crystal violet in 300mℓ of distilled water. Then 6mℓ of this staining solution was added into 1,000mℓ of cell suspension.

Sera samples and the antigen were brought to room temperature. Antigen solution 30μℓ was added to 40μℓ of each sample serum in a glass plate and then incubated for 8 min. at room temperature. Then the plate was hand rotated three times, at 4 and 8 min. after mixing and just before reading. Any sign of agglutination was considered positive [15].

7. TAT

The preparation of diagnostic antigen and procedure were conducted as described by Hur [16]. The prepared antigen was standardized according to the procedure of OIE, 2000 [14]. Briefly, inactivated S1119 whole cells were washed with 0.5 % phenol saline (0.85 %) and suspended in 0.5 % phenol saline (0.85 %) containing preservative, at

the concentration of 4.5 % (*v/v*). This concentrated antigen was diluted in phenol saline for use at 1 : 100 dilutions.

Serum samples and the antigen were brought to room temperature. Thereafter, quantities of 40 $\mu\ell$ of serum samples were placed in different tubes and mixed with 2 $m\ell$ of diluted antigen. The results were read after incubation at 37 ℃ for 48 hours. A positive reaction was one in which the serum mixture was clear and gentle shaking did not disrupt the flocculi. A negative reaction was one in which the serum antigen mixture was not clear and gentle shaking revealed on flocculi [17].

8. MET

The MET was performed as described by Alton et al [18]. Briefly, 0.1 M 2-mercaptoethanol(2-ME) solution in normal saline was made (sodium chloride, 8.5 g; 2-ME, 7.14$m\ell$ and distilled water to 1 liter) freshly and stored at 4℃. Test sera with a volume of 40$\mu\ell$ of each sample were placed in different test tubes and 1$m\ell$ of 0.1 M 2-ME in saline and 1$m\ell$ of concentrated TAT antigen diluted 1 : 50 in normal saline solution were added to each tube. The tubes were then shaken and incubated as described in TAT. The procedure of TAT was followed to interpret the titers obtained in the MET.

9. Statistical analysis

The seroprevalence was determined by considering the total number of animals tested and positive reactors using the formula given by Thrusfield [19]. The results were statistically analyzed for interpretation by using Chisquare tests (χ^2). Probabilities associated with the observed values of Chi-square, determined from relevant tables. Significance determined at 5% level. Odds ratio was obtained by the formula according to the Schlesselman [20] to find out the susceptibility of the species (sheep or goat) to brucellosis.

Results

1. Prevalence of brucellosis in sheep and goat

The overall prevalence of brucellosis in sheep and goat shown in Table 1, was 4.84 % (3 sheep) and 2.33% (7 goats) in among 62 sheep and 300 goats, respectively. In this study, it was found that the susceptibility of a goat to brucellosis was 2.13 times more than that of a sheep.

Table 1. Overall prevalence of brucellosis in sheep and goats

Species	Number of sera samples collected and tested	Number (%) of positive cases	Odds ratio
Sheep	62	3 (4.84)	1.00
Goat	300	7 (2.33)	2.13

2. Prevalence of brucellosis by various test

The prevalence of brucellosis was varied according to the serological tests carried out showed in Table 2. Among 362 small ruminants, irrespective of species (sheep or goat), the prevalence was highest in MET 2.76 % (n = 10), higher in TAT 2.21 % and lowest in both RBT and PAT 1.93 % (n = 7).

Table 2. Overall prevalence of brucellosis in small ruminants irrespective of species diagnosed by RBPT, PAT, TAT and MET

Number of sera samples collected and tested	Number of sera positive (%) by				Level of significance (χ^2-test)
	RBPT	PAT	TAT	MET	
362	7 (1.93)	7 (1.93)	8 (2.21)	10 (2.76)	*

*: Significant at 1% level of probability ($P<0.01$)

3. Age wise Brucella antibodies

Age wise prevalence of brucellosis presented in the Table 3 revealed that in case of both sheep and goats, 7 and 60 animals, respectively having 6 to 18 months of age, there was no positive reactor found but in case of age group of 19 to 30 months of 40 sheep, the prevalence of brucellosis were 2.5 % by RBT and PAT, and 5 % in TAT and MET. Besides, in case of 150 goats the prevalence of brucellosis was 1.33 % by RBT and TAT, and 2 % by PAT and MET. In case of age group of 31 to 42 months of 15 sheep, the prevalence of brucellosis were 6.67 % in RBT, PAT, and MET, but no positive reactor was found in TAT and at the same age group, in case of 90 goats, the prevalence of brucellosis was 3.33 % in RBT, 2.22 % in PAT and 4.44 % in TAT and MET. According to the tests performed, the prevalence of brucellosis in sheep and goat varied regarding various age groups. In both species, the more prevalence of brucellosis was found in case of aged animals (31 to 42 months). Statistically, the occurrence of brucellosis had no significant relationship with the age group.

Table 3. Age wise *Brucella* antibodies diagnosed by RBT, PAT, TAT and MET in sheep and goats

	Age (months) of animal	Number of sera					Level of significance (χ^2-test)
		Samples collected and tested	Positive (%) by				
			RBP	PAT	TAT	MET	
Sheep	6 – 18	7	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	NS*
	19 – 30	40	1 (2.50)	1 (2.50)	2 (5.00)	2 (5.00)	
	31 – 42	15	1 (6,67)	1 (6,67)	0 (0.00)	1 (6.67)	
Goats	6 – 18	60	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
	19 – 30	150	2 (1.33)	3 (2.00)	2 (1.33)	3 (2.00)	
	31 – 42	90	3 (3.33)	2 (2.22)	4 (4.44)	4 (4.44)	

*NS : Not significant

4. Area wise *Brucella* antibodies

The area wise prevalence of brucellosis has been shown in Table 4 and 5. The

prevalence of brucellosis in sheep in Bangladesh agricultural University Nutrition Farm was 4 % in both RBT and PAT, and 8 % in both TAT and MET. In case 18 sheep of Pharmacology Project, Department of Pharmacology, BAU, found no positive reactors in any of the performed tests.

Table 4. Area wise *Brucella* antibodies diagnosed by RBT, PAT, TAT and MET in sheep and goats

Species	Selected area	Number of sera					Level of significance (χ^2-test)
		Samples of tested	Positive (%) by				
			RBPT	PAT	TAT	MET	
Sheep	BAU nutrition farm	25	1 (4.00)	1 (4.00)	2 (8.00)	2 (8.00)	
	Pharmacology project	18	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
	BAU adjacent villages	7	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
	Villages−1*	12	1 (8.33)	1 (8.33)	0 (0.00)	1 (8.33)	NS***
Goats	Vet Clin, BAU	30	3 (10.00)	2 (6.67)	2 (6.67)	3 (10.00)	
	BAU adjacent villages	50	0 (0.00)	1 (2.00)	1 (2.00)	1 (2.00)	
	Villages−2**	190	2 (1.05)	3 (1.58)	3 (1.58)	3 (1.58)	
	Dhamrai Upazilla, Dhaka	30	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	

*Boyra and Char Nilaykha, and **Boyra, Char Nilaykha and Vangnamari, Ishwargonj Upazilla, ***Not significant

Table 5. Overall prevalence of brucellosis in small ruminants irrespective of species (sheep or goat) diagnosed by serological tests based on different districts

District	Sera tested	Positive cases number (%)
Dhaka	30	0 (0.00)
Mymensingh	332	10 (3.01)

No positive case was also found in 7 sheep of Bangladesh Agricultural University adjacent villages. Prevalence of brucellosis in 12 sheep of Ishwargonj upazilla was 8.33% by RBT, PAT and MET, 0.0 % by TAT. In case of total 300 goats, 30 goats were attended in Bangladesh Agricultural University Veterinary Clinic for treatment, tested for brucellosis and found 10%, 6.67 %, 6.67 % and 10 % prevalence of brucellosis by RBT, PAT, TAT and MET, respectively. In case of 50 goats of BAU adja-cent villages, the prevalence was 0.0%, 2.0%, 2.0% and 2.0% by the above tests respectively. 190 goats of villages of Ishwargonj upazilla showed the prevalence for brucellosis of 1.05 % by RBT, 1.58 % by PAT, TAT and MET and in case of 30 goats of Dhamrai upazilla it was 0.0 % in all serological tests. In this study, there existed a significant ($P<0.05$) association among different area and the prevalence of bru-cellosis

when the sera samples tested by RBT.

The prevalence of brucellosis in respect of pregnant and non-pregnant animals was presented in Tale 6. In this study, 5 sera samples from pregnant sheep and 57 sera samples from non-pregnant sheep were studied. In pregnant sheep, no positive case of brucellosis was found but in case of non-pregnant sheep, the prevalence of brucellosis was 3.5% in RBT, PAT, TAT, and 3.26% in MET. A total of 300 sera samples from goats were examined having 30 pregnant goats and 270 non-pregnant goats. In case of pregnant goats, the prevalence of brucellosis was 1.33% in all four serological tests and in case of non-pregnant goats it was 1.48%, 1.48%, 1.85% and 2.22% in RBT, PAT, TAT and MET, respectively.

Table 6. Prevalence of brucellosis in pregnant and non-pregnant sheep and goats*

Species	Criteria	Number of sera					Level of significance (χ^2-test)
		Samples	Positive (%) by				
			RBPT	PAT	TAT	MET	
Sheep	Pregnant	5	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	NS**
	Non-pregnant	57	2 (3.50)	2 (3.50)	2 (3.50)	3 (5.26)	
Goats	Pregnant	30	1 (3.33)	1 (3.33)	1 (3.33)	1 (3.33)	
	Non-pregnant	270	4 (1.48)	4 (1.48)	5 (1.85)	6 (2.22)	

*: Results based on RBPT, PAT, TAT and MET; **: Not Significant

Discussion

The main objective of this study was to determine the sero-prevalence of small ruminant brucellosis in Bangladesh. Brucellosis is a zoonotic disease [21]. Half a million new cases of brucellosis are reported world wide each year, but according to the World Health Organization (WHO), these numbers greatly underestimate the true prevalence. It is a systemic infection in which the bacteria initially localize in the regional lymph node, and then disseminate haematogenously to the organ of the reticuloendothelial system to multiply within phagocytic cells [22]. The release of bacterial endotoxin from phagocytic cells, produce the constitutional symptoms and sign of disease.

Brucellosis is a bacterial disease of world wide distribution and of major economic importance in most countries of the world, particularly amongst livestock industry. The prevalence varies considerably between herds or flock, between areas, and between countries, and details of the percentage of animals affected are of little for this reason [23]. The importance of brucellosis was primarily due to its public health significance and to the economic loss to the animal industry [21].

Bangladesh has been reported as an endemic area for brucellosis because a considerable number of human and animal populations are exposed to the infection each year [24, 25]. The present investigation revealed that the overall sero-prevalence of brucellosis in sheep

was 4.84% in sheep and 2.33% in goats. This is the first positive report of brucellosis in sheep in Bangladesh. Similar prevalence in sheep was reported by Sharma et al [26] and Bandey et al [27] in other countries. Abd-el-Ghani et al [28] found 2.97% brucellosis in ewes. Cerri et al [29] reported 2% brucellosis of goats. Rahman et al [10] reported 14.57% positive cases of brucellosis in caprine in different areas of Bangladesh. Radwan et al [30] found highest 11.6% brucellosis in small ruminants in Saudi Arabia, small ruminants raised on desert ranges found 0.5%, and small ruminants raised in small groups around individual homes are reported 0.4%. This difference might be due to that in Saudi Arabia, sheep and goat are reared commercially in farm, in dense populated herd. Bandey et al [27] found 3.2% *Brucella* positive Merino sheep in Kashmir valley. Hadad and Azawy [28] reported the prevalence was 5.5% in sheep and 5.3% in goats.

Rao et al [29] recorded the prevalence of brucellosis was 7% in goats of Andhra Pradesh. Burriel et al [30] found 16.8% of sheep and 13.1% of goats were positive to *Brucella* infection in Greece. Al-Majali [31] investigated the sero-prevalence of brucellosis in goats in Jordan and reported 27.7 % goats had antibodies against *Brucella*.

Species wise analysis of brucellosis revealed that, the risk of being infected by brucellosis of a goat is 2.13 times more than that of a sheep. Al-Izzi and Barhoom [32] found 23.3% sheep and 9.9 % goats were positive to brucellosis but Ismaily et al [33] revealed 0.9% of goats and 1.68% of sheep were positive in Sultanate of Oman. Hussain et al [34] reported that the percentage of brucellosis was relatively higher in ewes, 13% than buffalo (6.3%) and cattle (10.7%). Singh et al [35] reported that serologically, 0.6% of goats and 3.6% of sheep were found to be positive. Mrunalini and Ramasastry [36] reported that among the animals, most sero-positive reactions were in goats (7.0 %), followed by buffaloes (4.14%), cattle (3.8%) and sheep (3.3%). Ahl et al [37] reported that in case of goats and sheep of the flock, sero-prevalence of brucellosis was 11.3% for sheep and 2.5% for goats.

The study showed that the prevalence of *Brucella* antibodies in small ruminants was 1.93% with RBT and PAT, 2.21% with TAT and 2.76% with MET. Since there was statistically highly significant relationship between the prevalence of brucellosis determined by RBT and TAT, RBT and MET, PAT and TAT, and PAT and MET ($P<0.001$), the result of MET (2.76%) is considered to report the prevalence of brucellosis. Besides, MET has been found to be more sensitive and more specific test for detection of *Brucella* antibodies [38] and has been recommended to be stable and suitable test for routine diagnosis of brucellosis.

The higher prevalence in this study could be due to the more sensitive and more specific performance of TAT and MET that provides relatively accurate results.

The diagnosis of brucellosis is confirmed by isolation of *Brucella* by bacteriological culture or by the detection of an immune response by serological test to its antigens [39]. The diagnosis of brucellosis based exclusively on *Brucella* iso-

lation presents several drawbacks. The slow growth of Brucella may delay diagnosis for more than 7 days [40]. Also, the sensitivity is often low, ranging from 50 to 90% depending on disease stage, *Brucella* species, culture medium, quantity of bacteria and culture technique used [41].

The RBT is a screening test for serological diagnosis of *Brucella* infection and more sensitive than the CFT when testing culture positive animal [42]. The TAT is recommended for collection of quantitative information on immune responses, and is the most frequently used confirmatory serological test. In some countries *Brucella* positive serum samples are subjected to MET as confirmatory test [43]. The MET depends on ability of 2-ME to split the disulfide bonds in proteins. In the absence of urea the chemical selectively inactivate Ig M, leaving the Ig G intact.

In addition, TAT and MET has been found to detect antibodies in chronically infected animals while RBPT detects antibodies only in acutely infected subjects. As most of the previous investigators employed RBPT, SAT and CFT either individually or in combination, chronically infected animals might have escaped detection. This could be another factor responsible for the difference. But the prevalence recorded in this study with RBT (1.93%) is consistent with most of the previous reports[9]. It was reported that TAT is more sensitive than RBT [44] though Boargob and Muhammed [45] recommended the RBT as a single test for the detection of ovine and caprine brucellosis and Seddek [46] concluded that RBT and buffer acidified plate test (BAPAT) were effective and highly sensitive tests for detecting *Brucella* infection, whereas, TAT is of low sensitive and specificity.

Among the three age groups, the highest prevalence of brucellosis in both sheep (6.67%) and goats (4.44%) by MET, was found in 31 months to 42 months age group, though there was no significant association statistically between age group and the prevalence of brucellosis when sera samples were tested with RBT, PAT, TAT and MET. Sergeant [47] there was no apparent association between age and serological status, or age and the prevalence. Saleem et al [48] carried out an investigation to determine the seroprevalence of ovine brucellosis especially in herds that showed signs of abortion and reported a high prevalence rate was at 3 years of age, and was 56.6 % in ewes and 69.2% in rams but Ghani et al [49] stated that several epidemiological factors, such as age, sex, breed, lactation number, herd size and living conditions influence the seroprevalence of brucellosis. Mudit et al [50] stated that the brucellosis prevalence in goats varied with age as 1.63%, 0.58% and 1.65% of kids, young adults and adults, respectively.

The highest prevalence of brucellosis was found in Bangladesh Agricultural University Nutrition farm (8.0%) and Ishwarjong upazilla (8.33%) in case of sheep and in case of goats it was highest (2 %) in Veterinary Clinic, Bangladesh Agricultural University, attended for treatment. Statistically there existed no significant relationship between prevalence of

brucellosis and area. Siriwardane[51] showed the prevalence of brucellosis in Sri Lanka was varied from 1.2 to 20.0% in various areas. Kumar et al[52] reported that the prevalence of sheep brucellosis was higher in Punjab (50%) than in Rajasthan (32.73 %); and the prevalence of brucellosis in goats was highest in Rajasthan (29.7%) followed by Uttrar Pradesh (29.0%) and Punjab (15.8%). The prevalence of brucellosis may vary from flock to flock.

The higher prevalence of *Brucella* infection detected in the present investigation could be favored by the husbandry practices in the regions and the absence disease monitoring and control policy. The nomadic pastoralism prevailing in the study areas contributed well to the spread of infection from area to area and from flock to flock during mass movement of flocks and commingling at communal pastures and watering areas which supports the work of Richard[53]. Significantly higher prevalence rate (2.76 %) was found in Mymensingh district than the Dhaka district (0.0%). The difference in prevalence between the study areas is mainly due to difference in herding practices. In the Mymensingh district mixing of animals from various areas is common at communal grazing and watering areas. This variation in prevalence between the two regions could be supplemented by the presence of large number of goats, which are inherently more susceptible than sheep, in Mymensingh district than the Dhaka district. The demonstration of prevalence of brucellosis was varied and is depend on factors like climate, topography, and geographical localities, breed of animals and management practices[54]. The prevalence of caprine brucellosis were determined 23.64% at Modhupur in Tangail district, 31.82% at Bhaluka in Mymensingh district, 34.00% in Manikganj district and 16.66% in BAU campus and adjacent villages by PAT[10]. The difference of that might be due to the time passed, variation in methodology, sanitation and rearing system, keeping pattern, hygienic management, awareness of people, treatment of animals, improvement of veterinary services and reducing the number of domestic small ruminants specially sheep.

Brucellosis is an important disease with predilection for placenta and fetal membrane. The most common clinical features of brucellosis are placentitis and abortion[55]. In this study, the prevalence of brucellosis was higher in non-pregnant sheep but it was higher in pregnant goat. There was no statistical relationship between the pregnancy and the prevalence of brucellosis. Rahman et al[10] reported higher prevalence of brucellosis in goats with reproductive disorders but there was no report on association of brucellosis with pregnancy.

The over all prevalence of *Brucella* antibodies in small ruminants recorded in the present study was 2.76%. This indicates brucellosis is wide spread in Bangladesh. Since most of the human infections are caused by *B melitensis*, the most common *Brucella* of sheep and goats there is high risk for public health. Such findings of high prevalence in the absence of any vaccination against brucellosis indicate the occurrence of natural infection. It revealed the presence of pockets of infection in the Mymensingh in parti-

cular. Since there is close contact among pastoral community and livestock, higher prevalence of brucellosis is an indicator of potential risk for public health. Moreover, pastoral areas constitute an important source of small ruminants for national markets and can contribute to the spread of the infection to other areas and human beings. Further epidemiological studies and identification of the *Brucella* biotypes involved is recommended.

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