

First Report of Brucellosis in Dairy Cattle and Humans in Military Farms in Bangladesh

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Abstract

Brucellosis is a great concern for the dairy farming globally including Bangladesh. It is also an important zoonosis having public health significance. The seroprevalence, risk factors and species of *Brucella* prevalent in Bangladesh is known in large extent but the status of this disease in military farms is unknown. This study was performed to know the seroprevalence of brucellosis in 744 lactating cattle in eight military farms and 347 in contact humans using Rose Bengal Plate Test (RBT). The overall prevalence of bovine brucellosis was 2.3% (95% Confidence Interval [CI]: 1.4 - 3.7). The seroprevalence of bovine brucellosis varied from 0 - 3.6% in different farms. The overall seroprevalence of brucellosis was 0.7% (95% CI: 0.2 - 2.2) in humans in contact with military farms. The prevalence of bovine brucellosis varied in different military farm but the level is not high. The prevalence of brucellosis in accompanied humans is also very low. Further studies to identify possible factors responsible for the seroprevalence of brucellosis in humans and cattle in military farms are recommended.

Keywords: *Brucellosis; RBT; Seroprevalence; Military Dairy Farm; Bangladesh*

Introduction

Brucellosis is a major emerging zoonosis caused by the small, non-motile gram-negative and intracellular coccobacilli belonging to the genus *Brucella* [3]. It causes a great economic loss to the livestock industries through abortion, infertility, birth of weak and dead offspring, increased calving interval and reduction of milk yield [5]. In man, the clinical picture resembles many other febrile diseases, but sacroiliitis and hepato-splenomegaly are the most prominent. Brucellosis is endemic both for human and animal in Bangladesh and it is a notifiable disease in many countries but it is not a notifiable disease in Bangladesh [4,6-10]. The importance of brucellosis is not known precisely, but it can have a considerable impact on both human and animal health, as well as having socioeconomic effects, especially in areas where rural income relies largely on livestock breeding and dairy products. Brucellosis in human beings is caused by exposure to livestock and livestock products. Infection can result from direct contact with infected animals and can also be transmitted to consumers through raw milk and milk products [6].

The species of *Brucella* prevalent in humans and animals in Bangladesh has been reported [11-13]. However, these studies did not include cattle reared in military dairy farms in Bangladesh. There are eight military farms in various location of the country named as military farm Savar, Ishurdi, Lalmonirhat, Jossore, Chattogram, Comilla, Shornodip and Trishal, respectively. The total cattle population in these farms is more than 5000 which play a prime role to supply milk and dairy products to more than 1.5 lac defense personnel daily.

It is worthy to mention that around 2000 civilian work in these farms daily. The objective of this study is to report the seroprevalence of brucellosis in dairy cattle and in contact humans in military dairy farms in Bangladesh.

Materials and Methods

The study protocol of was peer reviewed and approved by the Ethical Review Committee of appropriate authority and informed written consent was taken from Remount Veterinary and Farm Directorate, Army Head Quarters, Bangladesh Army.

Collection and handling of blood samples and separation of sera

Patients with PUO (Pyrexia of unknown origin) were defined as those with body temperatures higher than 38°C on several occasions and lasting over a period of three weeks. A total of 437 patients were recruited from eight different military dairy farm workers. About 4 mL of blood was collected with disposable needles and Venoject tubes, labeled from human with the cooperation of a medical doctor, and transported to the laboratory on ice (after clotting) within 12h of collection.

About 5 - 7 ml of blood was collected from each of 744 dairy cows by jugular venipuncture following the method described above. Blood samples were kept in the refrigerator (2 - 8°C) in the laboratory and one day later sera were separated by centrifuging at 6000g for 10 minutes. Each serum was labeled to identify the human, animal and stored at -20°C. Each serum was divided into two tubes each containing about 1 - 1.5 ml of serum. One aliquot was used for testing and the other was preserved in a serum bank.

Serology

RBT was performed as described by following the appropriate technique [16]. Briefly, sufficient antigen, test sera, positive and negative control sera for a day’s testing were removed from refrigeration and brought to room temperature (22 ± 4°C). Equal volumes (30 µl) of serum and antigen (concentrated suspension of *B. abortus* biotype 1 (Instituto de Salud Tropical Universidad de Navarra, Spain) were mixed and rotated on a glass plate for 4 minutes. The result was considered positive when agglutination was noticeable after 4 minutes.

Statistical analysis

Descriptive statistics, 95% confidence interval of prevalence were performed in R 3.1.0 (The R foundation for Statistical Computing).

Results

A total of 744 sera samples were tested and the overall prevalence of bovine brucellosis was 2.3% (95% Confidence Interval [CI]: 1.4 - 3.7). The seroprevalence of bovine brucellosis varied in different farms. All tested cattle in Shornodip military farm were seronegative but the highest prevalence was found in Jossore military farm (3.6%). Table 1 describes the overall seroprevalence of bovine brucellosis and its distribution in different military farms. The seroprevalence of brucellosis was 0.7% (95% CI: 0.2 - 2.2) in humans in contact with military farms (Table 2). Figure 1 described RBT and agglutination reaction.

Military dairy farms	Tested	Positive	Prevalence (%)	95% Confidence Interval
Ishurdi	82	2	2.4	0.4 - 9.4
Lalmonirhat	102	2	1.9	0.3 - 7.6
Savar	91	2	2.2	0.4 - 8.5
Jossore	137	5	3.6	1.4 - 8.7
Chattogram	157	3	1.9	0.5 - 5.9
Comilla	65	2	3.1	0.5 - 1.2
Trishal	69	1	1.4	0.07 - 8.9
Shornodip	41	0	0	0 - 10.7
Total	744	17	2.3	1.4 - 3.7

Table 1: Seroprevalence of bovine brucellosis in different military dairy farms based on RBT, 2017 - 2018.

Tested	Positive	Prevalence (%)	95% Confidence Interval
437	3	0.7	0.2 - 2.2

Table 2: The seroprevalence of brucellosis in humans who worked in eight military dairy farms based on RBT test, 2017 - 2018.



Figure 1: Rose Bengal Test (left) reaction during RBT (no. 3, 5, 6 are positive and no. 1, 2, 4 are negative for brucellosis). Rose Bengal antigens used for diagnosis of brucellosis (right).

Discussion

The diagnosis of brucellosis is confirmed by isolation of *Brucella* species by bacteriological culture or by detection of an immune response to its antigens by serological tests. Diagnosis based exclusively on the isolation of *Brucella* species presents several drawbacks: the slow growth of *Brucella* may delay diagnosis by more than seven days, and sensitivity is often low, from 50 to 90 per cent depending on the diseases stage, the *Brucella* species, the quantity of bacteria, and the type of culture medium and technique employed and therefore the serological testing is important in the diagnosis of brucellosis [6].

The RBT is the main serological test used to diagnose *Brucella* species infection; sometimes it is more sensitive than the complement fixation test, especially in animals [6]. In this study for the first time describes the seroprevalence of brucellosis in dairy cattle and human in military dairy farms in Bangladesh by RBT. We observed 2.3% of bovine brucellosis in dairy cattle by RBT. Other authors from different parts of Bangladesh had also reported similar prevalence [1,7,15]. Higher seroprevalences [2,7] and lower seroprevalences [10,14] than our result had also been reported from Bangladesh. We observed 0.7% brucellosis in humans working in the military farms in Bangladesh. The apparent prevalence of human brucellosis reported from previous studies varied from 3.3% to 6.0% [4]. The true prevalence of human brucellosis was reported to be 1.1% [12]. One possible reason for such variation may be due to the difference in tests used and their interpretation. Interestingly none of the tested cattle under Shornodip military farm was positive for brucellosis. Being located in the Island it is easy to maintain good biosecurity practices which may not be possible in other places. Other than being located in the Island, the management practices in high prevalence and low prevalence farms can be compared to identify possible protective and risk factors for brucellosis in military farms in Bangladesh. The prevalence of bovine brucellosis varied in different military farm but the level is not

high. The prevalence of brucellosis in accompanied humans is also very low. Further studies to explore the possible factors responsible for the seroprevalence of brucellosis in humans and cattle in military farms are recommended.

Conclusion

The prevalence of brucellosis in cattle and accompanied humans is very low in military farms in Bangladesh and further studies to explore the possible factors responsible for the seroprevalence of brucellosis in humans and cattle are necessary.

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