SHORT COMMUNICATION

Seroprevalence of Bovine Brucellosis in Panchmahals and Mahisagar Districts of Gujarat State of India

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Abstract

Bovine brucellosis is a highly contagious zoonotic disease of word-wide importance both for livestock and human beings. This study aimed to assess the seroprevalence of bovine brucellosis in Gujarat's Panchmahals and Mahisagar districts. A total of 180 blood samples from bovine animals (90 cattle, 90 buffaloes) were collected and analyzed by Rose Bengal Plate Test (RBPT) and Indirect-Enzyme Linked Immunosorbent Assay (i-ELISA). The seroprevalence of brucellosis among cattle (n = 90) was 12.22% by RBPT and 4.44% by i-ELISA. In buffaloes (n = 90), the seroprevalence of brucellosis was 11.11% by RBPT and 0.00 % by i-ELISA. An overall seroprevalence observed was 11.67 and 2.22% by RBPT and i-ELISA, respectively. The seroprevalence of brucellosis in Panchmahals district (n = 90) was 7.78% by RBPT and 3.33% by i-ELISA, while in Mahisagar district, the seroprevalence was 15.56% by RBPT and 1.11% by i-ELISA. The findings suggest that there is a need to motivate and educate the dairy farmers through various extension programs regarding prevention and control aspects of brucellosis for reducing its prevalence in dairy animals.

Keywords: Bovine, Brucellosis, Gujarat, i-ELISA, RBPT, Sero-prevalence.

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Introduction

Brucellosis is one of the most ancient diseases first recorded in India in 1887 and has now become endemic throughout the country, with the prevalence of the disease ranging from 6.5 to 16.4 % in different species of livestock (Thoppil, 2000; Shome *et al.,* 2006; Kollannur *et al.,* 2007; Aulakh *et al.,* 2008; Jagapur *et al.,* 2013). The bacteria "*Brucella abortus*" are the leading cause of brucellosis in cattle. It is a highly contagious zoonosis caused by ingestion of contaminated unpasteurized milk & milk products, meat from infected animals, and handling aborted fetus, placenta, and other secretions with a bare hand. Inhalation of the organism is the most frequent route as an occupational hazard among herdsmen, dairy-farm workers, and workers in meat processing factories and in laboratory workers (Madkour, 2001). In India, brucellosis in livestock is responsible for a median loss of US $3.4 billion (Singh *et al.,* 2015). The disease in cattle and buffalo accounted for 95.6 % of the total losses occurring due to brucellosis in livestock populations. The disease is responsible for a loss of US $ 6.8 per cattle and US $ 18.2 per buffalo (Singh *et al.,* 2015). This study was undertaken in order to assess the seroprevalence of bovine brucellosis in the milk shed areas of Panchamahals and Mahisagar districts of middle Gujarat.

Materials and Methods

A total of 180 blood samples from bovine animals (90 cattle and 90 buffaloes) of selected dairy farmers were collected to study the prevalence of bovine brucellosis in Panchmahals and Mahisagar districts of middle Gujarat (India). From each dairy farmer, one animal was selected for the collection of blood samples based on the past history of abortion, repeat breeding, delayed postpartum estrus, or other infertility issues. Approximately 10 mL of blood sample was collected from the jugular vein of each animal using serum clot activator vacutainer. Samples were labeled using codes describing the species of animal. The clotted blood in the tubes was centrifuged at 2000 X g for 5 min to obtain clear serum and stored at -20°C in deep freezer until further laboratory analysis.

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Rose Bengal Plate Test (RBPT)

The antigen obtained from the Indian Veterinary Research Institute, Izatnagar, Uttar Pradesh, was used for Rose Bengal Plate Test. One drop (0.03 ml) of serum was taken on a glass slide by micropipette. The antigen and serum were mixed thoroughly with sterile toothpicks, and then the slide was rotated for four minutes, and the result was read immediately. Definite clumping/agglutination was considered a positive reaction, whereas no clumping/agglutination was considered negative.

Indirect-Enzyme Linked Immunosorbent Assay (i-ELISA)

For i-ELISA test, ID Screen® Brucellosis Serum Indirect Multi-Species for i-ELISA along with the user manual was procured from ID. Vet, Grabels, France. The test was performed as per the protocol outlined in the user manual. The optical density was measured at 450 nm, and the sample to the positive ratio (S/P%) was calculated for each sample by the following formula using sample and control values. Interpretation of result of i-ELISA was then made as per criteria mentioned in Table 1.

$$S/P\% = \frac{OD_{sample} - OD_{NC}}{OD_{PC} - OD_{NC}} \times 100$$

Where, S/P% = Sample to positive ratios, OD_{PC} = Optical density of positive control, and OD_{NC} = Optical density of negative control

The data obtained were analyzed using descriptive statistics to find out the overall, species-wise and district-wise percent prevalence of bovine brucellosis.

Results and Discussion

The seroprevalence of brucellosis among cattle was 12.22% by RBPT and 4.44% by i-ELISA (Table 2). In buffaloes, it was 11.11% by RBPT and 0.00% by i-ELISA. The overall seroprevalence of bovine brucellosis observed was 11.67% and 2.22% by RBPT and i-ELISA, respectively. The RBPT and i-ELISA test results were significantly different, the prevalence being higher with RBPT than i-ELISA ($p = 0.016$). A similar finding of 13.00 % seropositivity by RBPT was also reported by Trangadia *et al.* (2010) and Gogoi *et al.* (2017). However, a lower seroprevalence rate (~5%) by RBPT was reported by Swai and Schoonman (2010), Bhanu *et al.* (2013), and Shome *et al.* (2014), while a higher seroprevalence rate (30.40 %) was reported by Pathak *et al.* (2016) by RBPT. An overall seroprevalence of bovine brucellosis recorded by i-ELISA (2.22%) in the present study was lower than that (6.03 to 19.69%) reported by Bhanu *et al.* (2013), Shome *et al.* (2014), Gogoi *et al.* (2017), and Lindahl *et al.* (2019), while others reported a much higher prevalence of 22.18 to 41.55 % (Trangadia *et al.,* 2010; Jagapur *et al.*, 2013; Pathak *et al.,* 2016).

Further, in the present study, the seroprevalence of disease was somewhat higher in cattle than in buffaloes by RBPT, and none of the buffaloes tested by i-ELISA revealed positive results, even though 11.11% were positive by RBPT. The positive and negative results for RBPT and i-ELISA test detected are as shown in Fig. 1 and Fig. 2, respectively.

The seroprevalence of bovine brucellosis in Panchmahals district was 7.78 % by RBPT and 3.33 % by i-ELISA. In Mahisagar district, it was 15.56 % by RBPT and 1.11 % by i-ELISA. The prevalence of bovine brucellosis was somewhat higher in animals of Mahisagar district than in Panchamahals by RBPT (15.56 vs. 7.78%), while with i-ELISA the trend was reversed (1.11 vs. 3.33%, Table 2). The result of the prevalence of bovine brucellosis in both cattle and buffaloes in the study area suggest that there is a dire need to increase the knowledge level of dairy farmers about bovine brucellosis and especially

Table 1: Interpretation of result of i-ELISA Kit

Result interpretation	*Status*
S/P % ≤ 110%	Negative
110% < S/P % < 120%	Doubtful
S/P % ≥ 120%	Positive

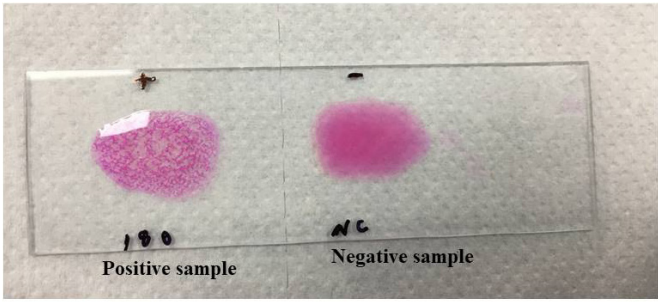

Fig. 1: Rose Bengal Plate Test

Table 2: Seroprevalence of bovine brucellosis using RBPT and i-ELISA techniques (n = 180)

			RBPT		*i-ELISA*	
Sr. no.	*Species and districts*	*No. of samples*	*No. positive*	*Prevalence (%)*	*No. positive*	*Prevalence (%)*
1	Cattle	90	11	12.22	4	4.44
2	Buffalo	90	10	11.11	0	0.00
Overall		180	21	11.67*	4	2.22
1	Panchmahals	90	7	7.78	3	3.33
2	Mahisagar	90	14	15.56	1	1.11
Overall		180	21	11.67*	4	2.22

Chi-square value between techniques = 5.833 *($p < 0.05$)

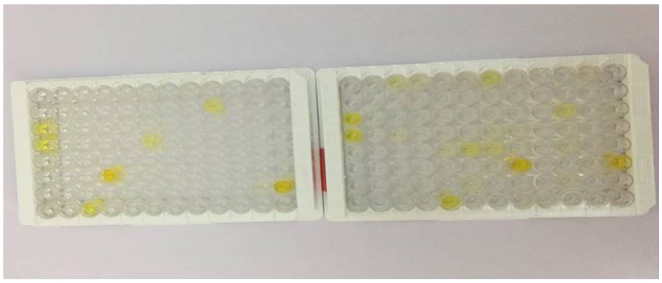

Fig. 2: Indirect-Enzyme Linked Immunosorbent Assay

its prevention and control aspects for reducing the prevalence of this devastating zoonotic disease of humans and animals. This can be done by organizing various extension activities to increase farmers' awareness of this disease and the adoption of preventive measures of bovine brucellosis. Jadav and Raval (2019) reported that majority of the dairy farmers (75.00 %) had a low level of knowledge about brucellosis disease.

Conclusion

Based on the present serological test, it was concluded that the prevalence of bovine brucellosis was 11.67% and 2.22% by RBPT and i-ELISA, respectively. The study recommends that a combination of RBPT and i-ELISA be successfully used to screen bovine brucellosis.

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