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Seroprevalence of Brucellosis Infection in Sheep and Goats of Raichur District in Karnataka

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Abstract: *Brucellosis is an important zoonotic disease globally that causes huge economic losses to the livestock owners and is of public health significance. Brucellosis in animals is endemic in India. In sheep and goats, Brucellosis is mainly caused by Brucella melitensis whereas Brucella ovis causes the disease in sheep. The symptoms in infected sheep and goats are abortions, stillbirths and the birth of weak offspring's. The present study was taken up to ascertain the sero-prevalence of brucellosis in sheep and goats of Raichur, district of the Hyderabad Karnataka region. A total of 300 serum samples from Raichur district were collected out of which 150 are from goats and remaining 150 from sheep were collected and subjected to Rose Bengal Plate test (RBPT), out of 300 sera samples from sheep and goats total 8 (2.66%) samples were found to be positive by RBPT. Where as prevalence was 2.00 % (3 out of 150) in sheep while it was showing 3.33% (5 out of 150) in goat. In conclusion, the finding of the study revealed that sero-prevalence of small ruminant Brucellosis in the study area was very low. However, the existence of the disease in the study area has possible risk of spread in the future and can pose great public health and economic threat.*

Keywords: *Brucellosis, sero-prevalence, sheep and goats and Karnataka.*

I. INTRODUCTION

Brucellosis is one of the world's major diseases. It is neglected zoonoses [1] largely due to lack of public awareness. Brucellosis causes huge economic losses to the livestock owners and is of public health significance. Brucellosis in animals is endemic in India. In sheep and goats, Brucellosis is mainly caused by *Brucella melitensis* whereas *Brucella ovis* causes the disease in sheep. Free grazing and movement with frequent mixing of flocks of sheep and goats also contribute to the wide distribution of brucellosis in these animals. The symptoms in infected sheep and goats are abortions, stillbirths and the birth of weak offsprings and metritis which may result in temporary infertility. Animals that abort may retain the placenta. Sheep and goats usually abort only once, but reinvasion of the uterus and shedding of organisms can occur during subsequent pregnancies. Others include drop in milk production due to the infection of the udder. Rams experience orchitis and epididymitis. In addition, animals with polyarthritis have been observed in endemic flocks [2]. Arthritis is seen occasionally in both sexes. Many non-pregnant sheep and goats remain asymptomatic.

Sero-prevalence studies form the backbone of epidemiology investigation and are used to identify herd infected with *Brucella* [3]. Serological evidence suggest that brucellosis is highly endemic in the most part of India [4], [5] and [6]. [7] first reported the isolation of *B. melitensis* from goat in India. Brucellosis is endemic in India and is prevalent in all parts of the country. Recent serological surveys of small ruminant Brucellosis indicated varying levels of infection in different states - 4.9% of sheep and 7.6% of goats in Karnataka [8], 11% sheep and 18% of goats in northern state of Delhi, 50% sheep and 16% goats in Punjab and 33% sheep and 30% goats in the western state of Rajasthan [9], 55% of goats in Andhra Pradesh [10] and 24% of goats and 4.7% of sheep in Uttar Pradesh [11], respectively. It was observed that flocks that had a history of abortion had high incidence of Brucellosis [10]. The tests commonly used for diagnosis of Brucellosis are the milk ring test, Rose Bengal Plate Test (RBPT), Standard Tube Agglutination Test (STAT), Microtiter Plate Agglutination Test (MAT) and ELISA. The RBPT is a rapid screening test for the diagnosis of Brucellosis. The sensitivity of RBPT is very high (>99%) but the specificity can be low and it could sometimes give a false positive result. RBPT test has proven useful as a herd screening test and is highly sensitive [12]. Therefore this study was undertaken with the main objective to do screening of sheep and goats in flocks from where abortions were noticed in the Raichur district.

II. MATERIALS AND METHODS

A. Study Area

The study was conducted in five talukas of Raichur district which comes under North-Eastern dry Zone of the Agro-climatic zone classified in the Karnataka state which lies in the 16.15° N of north latitude and 77.00° of east longitude. The climate remains almost dry throughout the year with mean monthly maximum temperature of 44.8° C in May and minimum of 16.6° C in December. The district receives very low and erratic precipitation with an average annual rainfall of 621 mm. Maximum rainfall occurs between June and September.

B. Study Population

There are about 281700 goats and 647379 sheep in Raichur district. The study population consisted of small ruminants kept under extensive management system. The animals under study comprised of local goat breeds and Kenguri sheep. Sheep and goats which were above 6 months of age, with no history of vaccination against brucellosis were included in the study. Individual animals age, species, sex category were recorded. Based on their sexual maturity animals were classified into ≤ 2 years, between 3-4 years and >4 years, respectively.

C. Study Design

A study was carried out to determine the sero-prevalence of Brucellosis in small ruminants kept under extensive management system. Randomization of sample was done by collecting samples from widely spaced villages and from different breeders and flocks of different taluka of Raichur district.

The present work on presence or absence of brucella antibodies in serum samples comprising goat sera (150) and sheep sera (150) collected from sheep and goats.

D. Sample Collection, Transportation and Storage:

About 5 ml of blood was collected aseptically from the jugular vein of individual animal in a vacutainer with serum clot activator. The vacutainers were kept in upright position at room temperature for about 2 hrs. The separated serum samples were collected and stored at -20°C till further use. These sera samples were tested for Brucella antibodies using RBPT, to detect Brucella antibodies in serum. Rose Bengal Plate Test was carried out by using rose Bengal plate test antigen.

E. Rose bengal plate test (RBPT)

The Rose Bengal Plate Test (RBPT) (Fig. 1) is often used as a rapid screening test [13] and considered as a reliable test in the diagnosis of brucellosis [14].

The sensitivity of RBPT is reported to be very high ($>99\%$) but the specificity can be disappointingly low. As a result, the positive predictive value of the test is low and a positive test result thus requires confirmation by a more specific test [15]. Gram negative bacteria such as *Yersinia enterocolitica*, *Vibrio cholera*, *Campylobacter fetus*, *Bordetella bronchiseptica* and *Salmonella* spp. may cross react with smooth *Brucella* spp. [16]. Despite the scanty and sometimes conflicting available information [17], [18], [19], [12], [20], [21] this test is internationally acknowledged as the test of choice for the screening of Brucellosis in small ruminants [22].

The RBPT antigen was procured from the Institute of Animal Health and Veterinary Biologicals (IAH and VB), Hebbal, Bangalore, Karnataka. All serum samples collected were screened using the RBPT. Briefly 30 μ l of stained rose Bengal antigen was dispensed on to card plate and then 75 μ l of sera samples were dropped alongside the stained rose Bengal Brucella antigen. By using the tip of the automatic micropipette tips, the sera were mixed and examined for agglutination. Positive and negative controls were employed for interpretation of the results.

Agglutinations were recorded as 0, +, ++ and +++ according to the degree of agglutination. A score of 0 indicates the absence of agglutination; + indicates barely visible agglutination; ++ indicates fine agglutination and +++ indicates coarse agglutination. Those samples with no agglutination (0) were recorded as negative while those with +, ++ and +++ were recorded as positive.

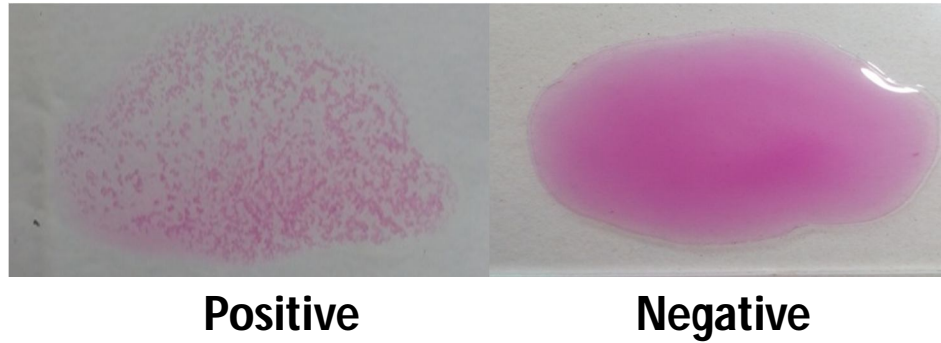


Fig. 1: Slide showing agglutination reaction on testing with Rose Bengal coloured antigen.

III. RESULTS AND DISCUSSION

Brucellosis is a worldwide zoonotic disease that is recognised as a major cause of heavy economic losses to the livestock industry and poses serious human health hazard. Several serological surveys of small ruminant Brucellosis indicated varying levels of infection in different states. Incidences of 4.9% in sheep and 7.6% in goats in Karnataka [8] 11% sheep and 18% in goats in Delhi, 50% sheep and 16% goats in Punjab and 33% sheep and 30% goats in the Rajasthan [9], 55% goats in Andhra Pradesh [10] and 24% goats and 4.7% sheep in Uttar Pradesh [11] have been recorded. In a national survey of sheep and goat Brucellosis, [23] tested serum samples, which included 6305 from sheep and 3849 from goats originating from 10 states. The cumulative incidence was 7.9% in sheep whereas in goats it was 2.2%. This survey indicated widespread prevalence of brucellosis in small ruminants in the country.

A total of 300 serum samples (150 from sheep and 150 from goats) were collected from small ruminants with no history of vaccination, kept under extensive management system. All serum samples were screened by RBPT. Overall prevalence of Brucellosis in small ruminants was 2.66 %. This result is in line with the report of [24] which report 1.99% Brucellosis using RBPT. Similarly [25], indicate the respective Brucellosis seroprevalence of 2.34% by RBPT. This level of sero-prevalence is in agreement with some of previous studies in Ethiopia. [26] in Jijiga, reported a sero-prevalence of 1.2% in sheep and 1.9% in goats; similar finding is reported by [27] which indicate a sero-prevalence of 1.6% in sheep and 1.7% in goats in Somali region. Likewise, [28] and [29] reported sheep Brucellosis of 1.5% and goat Brucellosis of 1.6%, respectively. On the contrary, it is lower than 4.6% and 9.7% of Brucellosis prevalence from other parts of the country by [30] and [27] respectively. This difference could be due to difference in agro-ecological location, sample size and animal management practices. Besides these, lack of hygienic measures against disease occurrence in the other study areas may contribute to the relatively higher prevalence of small ruminant Brucellosis. Similarly, the sero-prevalence of this study was relatively lower than that of [31] with respective prevalence 7.1 and 13.6% in sheep and goats. In addition, [27] reported sero-prevalence of 14.6% in sheep and 16.45% in goats in Afar region. This difference might be due to the variation in herding practices.

Table I Sero-Prevalence Of Brucellosis Based On Age And Sex In Sheep And Goat By Rbpt

Variables	Sheep						Goat					
	Age			Sex		Total	Age			Sex		Total
	<2 Y	>3 -4Y	>4 Y	Female	Male		<2Y	>3 -4Y	>4 Y	Female	Male	
No of sera samples tested	30	68	48	114	36	150	35	70	45	105	45	150
No samples positive by RBPT	0	2	1	2	1	3	1	3	1	4	1	5
%	0	2.94	2.08	1.75	2.77	2.00	2.85	4.28	2.22	3.8	2.22	3.33

Comparison of sero-prevalence of small ruminant Brucellosis was carried out for different species, animal origin, sex and age groups. The prevalence was 2.00 % in sheep while it was showing 3.33% in goat (Table 1). Likewise [32] explain the possible higher prevalence of Brucellosis in goats than in sheep is due to the greater susceptibility of goats to Brucella infection than sheep and the fact that unlike goats sheep do not excrete the Brucella organisms for longer periods of time which in turn can reduce the spread potential of the disease among sheep flock. The inconsistency among various studies could be due to the different sample proportion of sheep and goat which could affect the prevalence of the diseases.

Based on sex, the highest prevalence was found in male (2.77%) than female (1.75%) in sheep. Where as in goats highest prevalence was found in female (3.8 %) than male (2.22 %). When the sero-prevalence of small ruminant Brucellosis was compared between males and females, higher sero-positivity was observed in males over females in this study. This should not be overemphasized since very few numbers of positive cases observed which makes it difficult to make comparisons. However, [28] report significantly ($p < 0.05$) higher prevalence of Brucellosis in female sheep than in male sheep. . Similarly, it is an established fact that male animals are less susceptible to Brucella infection, due to the absence of erythritol. On the other hands, [33] and [25] have reported no observable difference in the prevalence of Brucellosis in male and female small ruminants similar to the present finding.

On the other hand a prevalence of 2.94 % was observed in the age group of 3-4 years and 2.85 % in younger animals (<2 years) (Table I). Higher sero-prevalence of small ruminant Brucellosis in the adult age groups than in the younger ones. Likewise, the work of [34] and [26] indicate the absence of significant variation in the seroprevalence of Brucellosis between different age groups of goats. However, [31] report the significantly ($p < 0.05$) higher sero-prevalence of Brucellosis in small ruminant more than 2 years than the other age categories. Similarly, it has been reported that sexually mature and pregnant animals are more prone to Brucella infection than sexually immature animals of either sex [32],[35]. On the other hand, younger animals are more resistant to infection and frequently clear an established infection, although latent infection can occur [36]. This may result from the fact that sex hormones and erythritol which stimulate the growth and multiplication of Brucella organisms tend to increase in concentration with the age and sexual maturity.

IV. CONCLUSION

In conclusion, the finding of the study revealed that sero-prevalence of small ruminant Brucellosis in the study area was very low. However, the existence of the disease in the study area has possible risk of spread in the future and can pose great public health and economic threat. Absence of strict cull and slaughter policy has compelled breeders to retain infected animals or sell to others as there is no compensation to farmers presently available if they decide to cull infected animals. To prevent this, regular screening of the newly introduced rams for crossbreeding should be carried out. Progress in control should be monitored serologically and evaluated epidemiologically. Veterinary extension should play a major role to guarantee the application of the sanitary procedures, improving the animal health biosecurity program and measures in rearing, raising and breeding places, education of personnel and dissemination of awareness as well as veterinary public health through various media. For successful commercial sheep and goat farming, control and eradication of brucellosis from sheep and goat population is of prime importance. Therefore, appropriate prevention and control method should be put in place to minimize the risk posed by this disease including elimination of sero-positive animal, vaccination and working on improving the awareness of the community toward the diseases.

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