

Short Communication

Co-Infection of *Mycobacterium avium* Subspecies *paratuberculosis* and *Brucella melitensis* in a Sirohi Breed of Goats in India

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ABSTRACT

Screening of goats (n=34) was performed using indigenously developed ELISA and SAT assays. Results revealed a total of 79.4% and 14.7% infection of caprine Johne's disease and caprine Brucellosis, respectively in a newly established goat herd at Popa Burj village in Mathura district of Uttar Pradesh, India. History indicated that the Sirohi breed of goats was recently purchased from the native tract in Sirohi district of Rajasthan. All of the five *Brucella* positive goats were found to be co-infected with Johne's disease. Study revealed the presence of two major infectious diseases in the herd of Sirohi goats, which demand immediate attention and need for developing control programmes for twin infections.

Key Words: *Brucella melitensis*, Johne's disease, Brucellosis

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Caprine Johne's diseases (CJD) and Caprine Brucellosis (CB) are not only economically important diseases of animals leading to production losses but are also zoonotic in nature and have public health significance. *Mycobacterium avium* subspecies *paratuberculosis* (MAP), the cause of CJD has wide host range and infects animals like cattle, buffaloes, goats, sheep, pigs, deer, rabbit, primates and human beings. Diarrhoea, weakness and low productivity are the main symptoms of CJD. MAP has been reported from all parts of the world (Barad et al., 2013, Braun et al., 1990, Robbi et al., 2002, Yadav et al., 2008, Mercier et al., 2010, Singh et al., 2013, Shroff et al., 2013). Brucellosis is predominantly caused by *Brucella melitensis* in goats and sheep and is major human infection (zoonosis) caused by consumption of raw milk and handling of meat and meat products of these animals (Xavier et al., 2010, Gupta, 2013). Indigenous ELISA kit (i-ELISA) and serum agglutination test (SAT) were used for the screening of CJD and CB, respectively. Farm located at Popa Burz (Raipura Jat) of Mathura district was recently established by purchasing Sirohi breed of goats from the native tract (Sirohi district) in Rajasthan. CB is an important disease of ruminants and about 18–40% of large ruminants reach abattoir due to infertility (Sharma et al., 1993). Beside its zoonotic implications, it is one of the major causes of abortions in goats and sheep. The study aimed to investigate presence of twin infections (CJD and CB) in a herd of Sirohi goats.

Thirty four (3 males and 31 females) goats of Sirohi breed were screened to estimate prevalence of CJD and CB in goat farm of Popa Burz (Raipura Jat) in Mathura district. The farm was recently established by purchasing Sirohi breed of goats from the native tract (Sirohi district) in Rajasthan. Animals were weak and suffering from diarrhoea. Serum samples were collected from all goats and screened for CJD and CB using i-ELISA and SAT, respectively.

Indigenous ELISA kit (i-ELISA) we have earlier developed for the screening of animals against JD (Singh et al., 2006) was used for screening. Protoplasmic Antigen (PPA) was harvested (Singh et al., 2008) from 'bison type' genotype of MAP isolated from a terminal case of JD in a Jamunapari goat (Singh et al., 2006). Whole cell sonicate was centrifuged and standardized at 0.1 ug per well of microtiter plate. Serum samples were used in 1:50 dilution and anti-goat horseradish peroxidase conjugate (Sigma Aldrich, USA) in 1:5000 dilution. Culture positive and negative samples of goats were used as positive and negative controls, respectively. Optical densities (OD) were transformed to S/P ratios and goats were negative (0.00–0.09), suspect (0.10–0.24), low positive (0.25–0.39), positive (0.40–0.99) and strong positive (1.0–10.00) for the status of CJD (Collins, 2002). Goats in strong positive and positive category were considered positive for CJD. Goats were screened by Standard Agglutination Test (SAT) following the procedure of Alton and colleagues (1988) with some modifications using *Brucella melitensis* plain antigen (Anonymous, 1971). The agglutination titres of $\geq 1:40$ were considered positive for brucellosis and those titres $\leq 1:20$ were taken as doubtful (suspected) and subjected to re-testing at monthly interval (Anonymous, 1971). Chi-Square test (X^2) applied to measure the significant association between the two tests (Siegel and Castellan, 1989).

Screening of 34 recently procured farm goats showed that, 27 (79.4%) were positive for CJD by i-ELISA and 5 (14.7%) for CB by SAT. Of 27 CJD positive samples, 4 (14.7%) were strong positive or super shedders. All 5 goats positive for CB were also positive for CJD (co-infected) (Table 1). Screening of 34 goats showed that, 5 (14.7%), 7 (20.5%), 22 (64.7%) and 0 (0.0%) were positive for co-infection of CJD and CB, negative for CJD and CB, positive for CJD and negative for CB and negative for CJD and positive for CB, respectively (Table 2). Results first time exhibited co-infection of MAP and *Brucella melitensis* in goats.

Table 1: Detection of MAP and *Brucella* infection in farm goats of Sirohi breed using Indigenous ELISA kit and SAT

MAP infection / Johne's disease		<i>Brucella</i> infection	
S/P ratio	Disease Status	n – Status in ELISA	n – Status in SAT
00.0–0.09	Negative	1 (2.9)	7 (20.5)
0.1–0.24	Suspected	3 (8.8)	29 (85.2)
0.25–0.39	Low positive	3 (8.8)	
0.4–0.9	Positive	23 (67.6)	5 (14.7)
1.0–10.0	Strong positive	4 (11.7)	
Total		34	34

*figures in parenthesis are percentage, n = number

Test	Combinations (n)			
	1	2	3	4
Indigenous ELISA	+	–	+	–
SAT	+	–	–	+
Total = 34	5 (14.7)	7 (20.5)	22 (64.7)	0 (0)

*figures in parenthesis are percentage, n= number

There is significant association between two tests with respect to Positive/Negative detection of cases ($P < 0.05$). Recently, commercial goat farming is growing at fast pace in India and Sirohi is very popular breed amongst goat farmers, therefore it is very important to know the incidence of chronic and incurable diseases like JD and brucellosis, in the native tract of this breed. Both these diseases may get distributed through the movement of the infected animals from the breeding tract to different parts of the country, as these herds form the foundation stock of many up-coming commercial goat farms. JD has been reported in goats worldwide with prevalence of 7.9, 76.9, 74.3 and 62.9% in Republic of Cyprus, USA, Chile and France, respectively (Liapi et al., 2011, Manning et al., 2002, Salgado et al., 2007, Mercier et al., 2010). In India, prevalence of CJD has been estimated were 78.0, 91.6 and 58.6% in Bhopal, Bareilly, and Mathura, respectively (Rajukumar et al., 2006, Tripathi et al., 2006, Kumar et al., 2007).

There are number of reports on the incidence of brucellosis in animal farms world-wide and incidence of brucellosis has been recorded as 4.4, 4.9, 0.03–0.1, 1.3, 4.9, 5.2–38.5, 32.8% from Iraq, India, France, C. Ethiopia, Egypt, Iran and Pakistan, respectively (Karim et al., 1979, Singh et al., 1998, Toma, 1990, Bekele and Kasali, 1990, Abdel Ghani et al., 1983, Zowghi and Ebadi, 1985 and Akhtar, 1992). Singh et al. (1998) also reported incidence of brucellosis in different breeds of goats in 7 district of UP and Punjab and was 4.0%. In another study, Singh et al. (2000) reported 2.7% incidence in goats through dot-ELISA (Singh et al., 2000).

Results showed high and moderate prevalence of CJD and CB in Sirohi breed of goats located in a farm herd at Mathura district. However, above prevalence indicate twin infections in native tract of Sirohi breed of goat, which is very popular in new commercial goat farms, therefore needs immediate attention for control.

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Table 2: Comparative status of co-infection of MAP and *Brucella* in Sirohi breed of goats at Popa Burz, Mathura using Indigenous ELISA and SAT

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