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Seroprevalence of bluetongue virus (BTV) antibodies in sheep and goats of semiarid Rajasthan

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ABSTRACT

The prevalence of BTV in field sheep and goats in semiarid districts of the Rajasthan state was studied. Sera samples (sheep, 1222; goats, 1158) were collected from selected 5 districts, of these 968 (sheep, 483, goats, 485) were randomly selected and tested by c-ELISA and analyzed. The overall prevalence of BTV in sheep was 36.02 % and in goats was 74.84 %. Combined overall prevalence in small ruminant population was 55.47 %. The present study revealed that prevalence of BTV is higher in local sheep and goats without clinical appearance of the disease in sheep, hence, there is possibility of circulation of same serotype of BTV in this area. The higher seroconversion in goats in contrast to sheep indicates the host preference of midges in day to day feeding.

Key words: Bluetongue virus, Goat, Sheep, Seroprevalence

As per 17th Indian Livestock Census Report (2003), small ruminant population in India is 18.5 crore that contribute substantially to national economy. In Rajasthan sheep (1 crore) and goats (1.68 crore) are generally reared through extensive system of rearing and one of major source of sustainable livelihood of rural poor. Among various economically important infectious diseases of small ruminants bluetongue is a non-contagious, viral disease caused by the *Orbivirus* genus of the family Reoviridae, and is transmitted by *Culicoides* midges.

In absence of an effective vaccine the control of spread of this virus lies on sound and extensive understanding of epizootiology and on availability of sensitive and reliable diagnostic tests to curtail the production losses not only in form of morbidity and mortality but also due to teratogenic effect of the virus (Srivastava *et al.* 1989). Several workers (Prasad and Srivastava.1995, Joshi *et al.* 1996, Sreenivasulu and Rao 1999, Nandi *et al.* 2005, Vegadabady *et al.* 2006) have documented disease incidence in different states. Dubey *et al.* (1988) and Prasad and Srivastava (1995) reported prevalence of this disease in Rajasthan but it was generally post incidence coverage confined to organized sheep farms and hence can not be taken as conclusive evidence of disease status in sheep reared under field conditions of a particular agroecological region. Present study was under mandate of the All India Network Project on BTD to assess current status of BTV in sheep and goat population of the state using more sensitive and reliable diagnostic test like ELISA.

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MATERIALS AND METHODS

Collection of sera samples: As per the agro-climatic distribution of the state 5 districts of semiarid zone i.e. Tonk, Ajmer, Jaipur/Sawai Madhopur, Bhilwara and Bundi, were selected for collection of sheep and goats sera samples during the period 2003-04. Samples (sheep, 1222; goats, 1158) were collected from different villages of 5 districts using sterile plastic syringes. Samples were transported to laboratory after clotting under cold chain. Serum was separated aseptically in sterile plastic vials and stored at –20°C until use. Of these 968 sera samples (sheep, 483; goats, 485) were randomly selected for testing (Table 1).

Competitive ELISA: The test was performed as per Afshar *et al.* (1987), with some modifications by using ELISA kits. Antigen coated wells were rinsed twice with washing buffer and the controls and serum samples in 50 µl quantities were added in duplicates at a dilution of 1:5 followed by the addition of same quantity of BTV group specific MAb diluted at 1:100 except diluent wells. The plates were shaken well for mixing and incubated 2h at room temperature (22°–28°C). The wells were washed thrice and 100µl of peroxidase conjugated anti-mouse immunoglobulin diluted to 1:300 was added to each well and incubated for 1 h at room temperature in dark. After washing the plates for 5-times 100µl substrate–OPD was added to each well and incubated for 10 min in dark. The reaction was stopped by the addition of stopping

buffer and the OD was recorded at 492nm on ELISA reader. The different controls included diluent, weak positive, strong positive and negative serum. The percent inhibition (PI) was calculated as per the formula given with the kit with the help of computer.

$$PI = \frac{\text{Adjusted average OD of test sample}}{\text{Adjusted average OD of negative serum}} \times 100$$

where, adjusted OD = Average OD of the serum-Average OD of the diluent.

The sera samples having the PI values <50% were classified as negative, 50-80 % as weak positive and 80 % and above were strong positive.

RESULTS AND DISCUSSION

The prevalence of BTV antibodies in sheep for the 5 districts (Table 1) revealed higher seroconversion in Jaipur/ Sawai Madhopur districts (44.68%) followed by Ajmer (44.08%), Tonk (39.52%), and Bhilwara (28.91%) and lowest in Bundi (24.03%). Irrespective of breeds the overall prevalence for BTV in local sheep of semiarid Rajasthan was 36.02 %, which is quite higher in comparison to AGID test based as reported 1.2% by Dubey *et al.* (1988) in flocks under ORP villages. The higher seroconversion in sheep observed in this study was in accordance with the findings reported by Das *et al.* (1997), in farm sheep (more than 60%) and Nandi *et al.* (2005) in indigenous sheep of Gujarat (52.77%) using ELISA system.

In goats the highest prevalence of BTV was recorded in Bundi and Jaipur/Sawai Madhopur districts (82.60%) followed by Bhilwara (73.14%), Tonk (67.32%) and lowest in Ajmer (65.2%). The overall prevalence of BTV was 74.84%, which is higher than the earlier AGID based observation (2.67% and 2.63%) of Dubey *et al.* (1988) and Doddamani and Hari Babu (2007) in village flocks of goat in Rajasthan and Karnataka respectively. The prevalence recorded in present study was in agreement with the findings reported by Prasad and Srivastava (1995) using ELISA based test to a tune of 23.33% in Rajasthan, 36.50% in Haryana and 39.39% in Himachal Pradesh and by Barbuddhe *et al.* (2005) among goats in Goa (58.01%).

The overall prevalence in sheep and goat revealed that 537 out of 968 sera were positive for BTV antibodies (55.47%), which is higher than the 9.1% overall observation recorded by Dubey *et al.* (1988).

Further, higher seroconversion in goats than sheep is another nitch in the epidemiology of the BTD. In semi arid regions of Rajasthan the sheep and goats are reared together in the same coral or in neighborhood under similar extensive system of management (Mann *et al.* 1980). Low seroconversion in sheep than that in goats in present study appeared to be direct evidence of preferential feeding of *Culicoides* midges. Such a preference was noticed and reported for cattle but not for goats (Radostits *et al.* 2000).

From the present study it can be concluded that though there has been no report of active clinical disease in Rajasthan state the presence of BTV antibodies in majority of the population indicates perhaps circulation of same serotype of virus, which is unable to produce clinical disease in pre-exposed animal population and hence, there is an urgent need of serotyping of BTV circulating in this area. The higher seroconversion in goats in contest to sheep indicates the host preference of midges in day to day feeding; therefore an attempt to keep goats with sheep flocks in endemic area if practiced may protect the sheep from excess exposure.

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Table 1. Bluetongue disease testing by c-ELISA

Districts	Samples collected		Samples tested		BTV positive		% positivity	
	Sheep	Goat	Sheep	Goat	Sheep	Goat	Sheep	Goat
Tonk	423	238	109	101	42	68	39.52	67.32
Ajmer	273	174	93	69	41	45	44.08	65.21
Jaipur/S.MP	129	157	94	92	42	76	44.68	82.60
Bhilwara	290	291	83	108	24	79	28.91	73.14
Bundi	107	298	104	115	25	95	24.03	82.60
Total	1,222	1,158	483	485	174	363	36.02	74.84
Total (sheep + goats)	2,380		968		537		55.47	

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