Original Research

Seroprevalence of Bovine Herpes Virus-1 among Cattle and Buffaloes in Central Kerala, India

P. V. Tresamol, K. Mery Rincy and P. Amel Dev

Department of Veterinary Epidemiology and Preventive Medicine, College of Veterinary and Animal Sciences, Mannuthy, Kerala, INDIA

***Corresponding author:** pvtresamol@yahoo.co.in

Rec. Date:	Jun 11, 2019 09:52
Accept Date:	Sep 03, 2019 05:39
DOI	10.5455/ijlr.20190611095243

Abstract

A cross-sectional study was conducted during 2016-17, for assessing the seroprevalence of Bovine Herpes Virus-1 among cattle and buffaloes maintained in organised and unorganised farms in Thrissur and Palakkad districts of Kerala, India. A total of 600 animals including 517 cattle and 83 buffaloes were included in the study. The sera samples from the animals were subjected to Avidin Biotin Enzyme Linked Immunosorbent Assay (AB-ELISA) for detection of antibodies to BHV-1. Screening of serum samples from 600 animals showed an overall positive reaction of 12.0 per cent. Among 517 cattle including 21 vechur cows 55 animals including 3 Vechur cows showed positive reaction (10.63 per cent). Among 83 buffaloes 17 (20.48) were positive for antibodies to BHV-1. Even though the rate of prevalence was low, the detection of antibodies to BHV-1 clearly demonstrated the establishment of infection among cattle and buffaloes in central Kerala.

Key words: BHV-1, Bovines, ELISA, Infectious Bovine Rhinotracheitis, Seroprevalence

How to cite: Tresamol, P. V., Rincy, K. M., & Dev, P. A. (2019). Seroprevalence of Bovine Herpes Virus-1 among Cattle and Buffaloes in Central Kerala, India. *International Journal of Livestock Research, 1*(10), 68-73. doi: 10.5455/ijlr.20190611095243

Introduction

Infectious bovine rhinotracheitis (IBR) caused by the bovine herpes virus-1 (BHV-1) is a highly contagious disease of bovines. It is an economically important disease affecting mainly cattle and buffaloes and is characterised by respiratory and genital tract infections including keratoconjunctivitis, pustular vulvovaginitis, balanoposthitis, abortions, infertility and meningoencephalitis (Saminathan *et al.*, 2016). Infectious bovine rhinotracheitis is one of the important respiratory/ reproductive viral diseases of bovines in India (Kiran *et al.*, 2005). The disease was first reported in India in 1976 (Mehrotra *et al.*, 1976) and since then seroprevalence of BHV-1 has been reported from different states of India. Nandi *et al.*, (2011)

reported highly variable seroprevalence of BHV-1, ranging from zero percent to 71.1 per cent in different parts of India. Kollannur *et al.*, (2014) showed an overall seropositivity of 32.25 per cent among cattle from different states with highest in Uttar Pradesh and lowest in Himachal Pradesh.

Bovine Herpes Virus-1 is a member of the the genus Varicellovirus in the subfamily Alphaherpesvirinae of family Herpesviridae. Three subtypes of BHV-1 are identified worldwide such as BHV-1.1, BHV-1.2a and BHV- 1.2b (Biswas *et al.*, 2013). It infects cells of the upper respiratory tract and leads to rhinitis, conjunctivitis, and tracheitis. Concurrent bacterial infection may lead to bronchopneumonia by reducing lung clearance mechanisms and inducing immunosuppression. Infection of non-immune pregnant cows results in systemic infection, foetal infection and abortion. Infectious bovine rhinotracheitis is the commonest form of BHV-1 infection and is characterized by respiratory symptoms like fever, coughing, increased respiratory rate and nasal and ocular discharge along with systemic signs such as anorexia, depression and decreased milk production (Straub, 1991). Genital infections lead to pustular vulvovaginitis in females and balanoposthitis in males. Abortion is sequelae of a respiratory form of infection. BHV-1 crosses the maternal foetal barrier to produce infection to the foetus. Absence of vaccination, intensive rearing, purchase and mixing of animals without screening and practice of natural insemination with unscreened bulls are the important predisposing factors causing the infection.

The sources of infection include the nasal exudates and the respiratory droplets, genital secretions, semen, foetal fluids and tissues of the infected animals. Transmission occurs mainly through direct contact with infected animals, aerosol route and virus-contaminated semen from infected bulls. The BHV-1 virus can become latent following a primary infection with a field isolate or vaccination with an attenuated strain. Latent and the subclinical infections are common in IBR (Ranganatha *et al.*, 2013) which can be identified through the detection of antibodies against BHV-1 in serum (Lemaire *et al.*, 2000). Several factors can cause reactivation and excretion of latent virus, which results in maintenance of BHV-1 within a herd (Muylkens *et al.*, 2007). Diagnosis of BHV-1 infections can be done by virus isolation, detection of viral antigens or by detection of genomic DNA by polymerase chain reaction (PCR). The molecular technique like PCR is considered as an excellent tool for the fast and very sensitive detection of viral genomes in biological and clinical specimens. However, enzyme-linked immunosorbent assays are used as a method of choice for routine diagnosis of BHV-1-infected animals. Indirect ELISA had been widely used for assessing the seropositivity of IBR among farm animals. The present study records the current status on seroprevalence of infectious bovine rhinotracheitis among bovines of central Kerala based on avidin-biotin ELISA(A-B ELISA).

DOI 10.5455/ijlr.20190611095243

Materials and Methods

A cross-sectional study was conducted during 2016-17 among cattle and buffaloes maintained in organised and unorganised farms in Thrissur and Palghat districts of Kerala. A total of 600 animals including 517 cattle and 83 buffaloes were included in the study. Blood samples were collected from the animals, sera were separated and inactivated at 56°C for 30 minutes and preserved at –20°C until use. The sera samples were subjected to A-B ELISA for detection of antibodies to BHV-1 using the kit developed at National Institute of Veterinary Epidemiology and Disease Information (NIVEDI), Bangaluru. The ELISA procedure was performed as per the manufacturer's protocol. Serum samples including control and test sera were diluted in blocking buffer (1:100) supplied. Hundred microlitre of the diluted control and test sera were dispensed to the BHV-1 Ag coated ELISA plates and incubated on the shaker at 370C for one hour. The wells were then washed three times using washing buffer. Then 100 µ of the Biotin-Anti IgG conjugate (1:10,000) was dispensed to all wells and incubated on the shaker at 370C for one hour. Repeated the washing procedure and dispensed 100 µ of the Avidin-HRPO conjugate (1:10,000) to all wells and incubated on the shaker at 370C for 20 minutes. After washing, 100 µ of the chromogen-substrate solution was added to all wells and incubated on the shaker at 370C for 10 to 12 minutes. The enzyme substrate reaction was stopped by adding 50 µ of stopping solution (1M H2SO4) to all wells. The absorbance values (OD) were recorded at 492 nm in the ELISA reader. Interpretation of the results was done as follows. Test Samples were considered positive if the OD Values were greater than 'X' where X = Average OD of Strong Positive x 0.64

Results and Discussion

Out of 600 serum samples tested using AB ELISA, 72 samples showed positive reaction (12.0 percent). Among 517 cattle, 55 animals showed positive reaction (10.63 per cent).Out of 517 cattle 21 were Vechur cows, among which three cows showed positive reaction. In this study, AB ELISA has been employed to screen the serum samples since the test is well suited for screening and analysis of a large number of samples as suggested by Salas *et al*. (2013). The results of the present study is similar to the observations made by Rajesh *et al*. (2003), who reported a seroprevalence of 14.88 among dairy cattle in Kerala whereas Kollannur *et al*., (2014) reported a higher seroprevalence of 22.5 percent. The previous studies reported a higher seroprevalence among cattle as 36.31 per cent in Gujarat (Kathiriya *et al*., 2018), 34.34 per cent in Chattisgarh, 36.51 per cent in Punjab, 55.42 per cent in Uttar Pradesh and 40.71 per cent in Uttarakhand (Kollannur *et al*., 2014). Seropositivity of 10.63 per cent clearly suggests the establishment of infection among cattle in central Kerala, as IBR vaccinations are not practiced in these regions. In the absence of vaccination, presence of antibodies can only be caused by exposure to the pathogen (Kampa *et al*., 2004).

DOI 10.5455/ijlr.20190611095243

The present intensive management system with high density of cows might have promoted viral spread and increased contact of healthy susceptible animals with infected animals as suggested by Chandranaik *et al.* (2014).

Table 1: Seroprevalence of BHV-1 among ruminants

Sl.No.	Animals	Number tested	Number positive	Per cent
1	Cattle	517	55	10.63
2	Buffaloes	83	17	20.48
	Total	**600**	**72**	**12.0**

Chi square value = 6.562; p-value = 0.0104; Significant difference between two proportions at 0.05 levels*

Screening of serum samples from 87 buffaloes showed positive reaction in 17 (20.48 per cent) animals. . Previous studies in India reported a higher seroprevalence of IBR among buffaloes as 33.99 per cent in Gujarat (Kathiriya *et al*., 2018), 35.28 per cent in Utter Pradesh (Verma *et al*.,2014) and 52.5 per cent in South Indian states (Renukaradhya *et al*., 1996). A low prevalence in the present study might be due to a small sample size and different geographical locations. A significant high proportion of seroprevalence was observed in buffaloes when compared to cattle in this study. The variations in the prevalence of the disease might be due the factors such as sample of population screened, intensity of dairy farming, inter-mixing of animals, unrestricted movement of infected animals and the extend of control measures adopted.

The factors responsible for occurrence of antibodies to BHV-1 in cattle and buffaloes might be due to use of contaminated semen for insemination, intensive rearing practices or introduction of animals to the farms without screening. The purchase of animals from infected farms for replacement will increase the risk of infection and higher seroprevalence. Seropositive animals may also occur due to virus latency, which is an inherent characteristic of the BHV-1 (Chandranaik *et al*., 2014).

Currently no vaccination is practised in Kerala against BHV-1 infection. Since none of the animals included in the study were vaccinated against BHV-1, the seroprevalence in these animals clearly indicated exposure to the virus as suggested by Kampa *et al.* (2004). Nandi *et al.*, (2011) reported highly variable prevalence rates of IBR in different parts of India, and suggested that these variations may be attributed to differences in husbandry and management practices or geographical differences. There were no specific clinical signs suggestive of BHV-I infection in screened animals except for history of abortion in five animals. Pritchard *et al*. (2003) also reported absence of clinical signs in a herd with over 70 per cent of seropositive animals to BHV-1. Geraghty and O'Grady (2012) reported a significant association between serological evidence of exposure to BHV-1 and reduced conception rate in a study of eight commercial dairy herds in Ireland. In contrast, Waldner (2005) could not find any evidence of association between BHV-1 serological status and reproductive performance in a large study of Canadian beef herds. Patil *et al.* (2017) reported higher

 DOI 10.5455/ijlr.20190611095243

seropositivity of BHV-1 in cases of abortion, metritis, repeat breeding and retention of placenta in organized dairy farms of India.

Conclusion

Seroprevalence of BHV-1 among ruminants in Kerala warrants the need of an intensive control and surveillance program for reducing the infection rates. Vaccination of Cattle and buffalo population in this area can be considered along with sero-monitoring for control of the disease. In addition regular attempts to isolate and characterize the causative agent for IBR has to be attempted. Inactivated and attenuated live vaccines are available for IBR, which is useful for protecting from clinical disease and markedly reducing the subsequent shedding of field virus. Although vaccination may not prevent field virus infection of individual animals, spreading of wild-type virus in infected herds is efficiently reduced.

References

1. Biswas S, Bandyopadhyay S, Dimri U, Patra P.H.2013. Bovine herpesvirus-1 (BHV-1) - A re-emerging concern in livestock-A revisit to its biology, epidemiology, diagnosis, and prophylaxis. Vet. Q. 33:68–81.
2. Chandranaik, B.M., Rathnamma, D., Patil, S.S., Ranganatha, S., Kovi, R.C., Akhila, D.S., Isloor, S., Renukaprasad, C. and Prabhudas, K. 2014. Cloning of partial glycoprotein B gene and molecular epidemiological studies of bovine herpes virus -1 isolates. Indian J. Anim. Sci., 84: 108–114.
3. Geraghty, T. and O'Grady, L. 2012. Effect of exposure to bovine herpes virus 1 on conception rate: a multivariate analysis. Proceedings of 27th World Buiatrics Congress, Lisbon, Portugal. pp. 205–206.
4. Kampa, J., Stahl, K., Lopez, M.J., Chanlun, A., Aiumlamai, S. and Alenius, S. 2004. BVDV and BoHV-1 infections in dairy herds in Northern and Northeastern Thailand. Acta Vet. Scand., 45: 181-192.
5. Kathiriya J., Sindhi, S., Mathapati, B. and Bhedi, K.2018.Seroprevalence of Infectious Bovine Rhinotracheitis (BHV-1) in dairy animals with reproductive disorders in Saurashtra of Gujarat, India. Inter. J. Curr. Microbiol. Appli. Sci. 7: 1371-1376
6. Kiran, K. K., Ravi, P. and Prabhudas, K. 2005. Infectious bovine rhnotracheitis- National survey of IBR antibodies by AB-ELISA kit. Annual Report of Project Directorate on Animal Disease Monitoring and Surveillance, ICAR, Bangalore 7-10.
7. Kollannur, J. Davis., Radhika, S. and Chauhan, R.S. 2014. Epidemiological studies on infectious bovine rhinotracheitis (IBR) in different parts of India. Int. J. Livest. Res., 4: DOI 10.5455/ijlr.20140717052204.
8. Lemaire, M., Meyer, G., Baranowski, E., Schynts, F., Wellemans, G., Kerkhofs, P. and Thiry, E. 2000. Production of bovine herpesvirus type 1-seronegative latent carriers by administration of a live-attenuated vaccine in passively immunized calves. J. Clin. Microbiol., 8: 4233-4238.
9. Mehrotra, M.L., Rajya, B.S. and Kumar, S. 1976. Infectious bovine rhinotracheitis (IBR) kerato-conjuctivitis in calves. Indian J. Vet.Pathol., 1: 70-73.
10. Muylkens, B., Thiry, J.,Kirten, P., Schynts, F. and Thiry, E.2007.Bovine herpes virus-1 and Infectious bovine rhinotracheitis. Vet.Res.38:181-209.
11. Nandi, S., Kumar, M., Yadav, V. and Chander, V. 2011.Serological evidences of Bovine Herpesvirus-1 infection in bovines of organized farms in India. Transbound.Emerg. Dis., 58: 105-109.

12. Patil,S.S., Prajapati,A., Krishnamoorthy, P., Desai G.S., Reddy, G.B.M. Suresh K.P. and Rahman. H. 2017. Seroprevalence of infectious bovine rhinotracheitis in organized dairy farms of India. Indian J. Anim. Res., 51 : 151-154.
13. Pritchard, G. C., Banks, M. and Vernon, R. E. 2003. Subclinical Breakdown with Infectious Bovine Rhinotracheitis Virus Infection in Dairy Herd of High Health Status. Vet. Rec.,153 113-117.
14. Rajesh, J. B., Tresamol, P. V. and Saseendranath, M. R. 2003. Seroprevalence of infectious bovine rhinotracheitis in cattle population of Kerala. Indian Vet.J. 80: 393-396
15. Ranganatha, S., Rathnamma, D., Patil, S.S., Chandranaik, B.M., Isloor, S., Veeregowda, B.M., Narayanabhat, M. and Srikala. 2013. Isolation and molecular characterization of bovine herpes virus-1 by polymerase chain reaction. Indian J. Anim. Res., 47: 340-343.
16. Renukaradhya, G.J., Rajasekhar, M. and Raghavan, R. 1996. Prevalence of infectious bovine rhinotracheitis in southern India. Rev Sci Tech. 15:1021-8.
17. Salas, R. D., Aguirre, A.C., Palacios, M. F., Vazquez, Z.G., Romero, C.A. and Dominguez, A.M. 2013.Seroprevalence and risk factors associated with infectious bovine rhinotracheitis in unvaccinated cattle in southern Veracruz, Mexico. Afr. J. Microbiol. Res., 7: 1716-1722.
18. Saminathan M , Rana R , Ramakrishnan M A , Karthik K , Malik YS and Dhama K. 2016 Prevalence, diagnosis, management and control of important diseases of ruminants with special reference to Indian scenario. J. Exper. Biol. Agri. Sci. 4(3S): 338-367
19. Samrath, D., Shakya, S., Rawat, N., Rani, V. and Singh, F. (2016). Isolation and adaptation of bovine herpes virus Type 1 in embryonated chicken eggs and in Madin–Darby bovine kidney cell line. Veterinary World. 9:222-225.
20. Straub O C. 1991. BHV1 infections: relevance and spread in Europe. Comp. Immunol. Microbiol. Infect. Dis. 14:175–186.
21. Verma, A.K., Kumar, A., Sahzad, Reddy, N.C. and Shende, A.N.2014. Seroprevalence of infectious bovine rhinotracheitis in dairy animals with reproductive disorders in Uttar Pradesh, India. Pak. J. Biol. Sci. 17:720-724.
22. Waldner CL. Serological status for N.caninum, bovine viral diarrhoea virus, and infectious bovine rhinotracheitis virus at pregnancy testing and reproductive performance in beef cattle. Anl. Reprod. Sci. 2005; 90:219–242. doi: 10.1016/j.anireprosci.2005.03.017.

DOI 10.5455/ijlr.20190611095243