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Seroprevalence to bovine herpesvirus type 1 in sheep in Turkey

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ABSTRACT

In the present study, sera from 1146 sheep from eight different locations in the northern provinces of Turkey were investigated against bovine herpesvirus type 1 (BHV-1) by a conventional method, as serum neutralization. Prevalence of antibodies detected against BHV-1 in sheep was found to be 1.74% (20/1146). Serum samples obtained from sheep from 4 different provinces were detected negative against BHV-1. As a result, with this study presence of BHV-1 was detected for the first time in sheep in the northern provinces of Turkey.

Key words: sheep, bovine herpes virus-1, neutralization, seroprevalence, antibody, Turkey

Introduction

Bovine herpesvirus-1 (BHV-1) is a member of the *Varicellovirus* genus, *Alphaherpesvirinae* subfamily, *Herpesviridae* family and has a positive sense double-stranded DNA genome (ALY et al., 2003; MWEENE et al., 2003). The virus creates different infections in ruminants associated with respiratory symptoms such as infectious bovine rhinotracheitis (IBR), genital infection as pustular vulvovaginitis (IPV) and balanoposthitis (IBP) in bulls, conjunctivitis, encephalitis, abortions and fatal multi-systemic infections (MWEENE et al., 2003). BHV-1 can establish latent infections in cattle and this fact serves as a constant source of infection during viral reactivation and re-excretion periods. The major economic losses in the cattle industry occur due to respiratory and reproductive disease caused by this virus (CASTRUCCI et al., 1997).

BHV-1 is currently widespread all over the world, for example the USA, Canada, Zaire, Italy, Belgium, India and Turkey (CASTRUCCI et al., 1997; BILGE DAGALP, 1998;

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BOELAERT et al., 2000; YESILBAG et al., 2003; RAJKHOWA et al., 2004). The BHV-1 serological surveys with cattle carried out in Turkey ascertained that this virus spreads among dairy and beef cattle in many regions of the country (CABALAR and AKCA, 1994; ALKAN et al., 2000; ALY et al., 2003; AKCA et al., 2004). Previous research by CABALAR and CAN-SAHNA (2000) revealed that the seropositivity rate of BHV-1 was between 20 and 74%.

BHV-1 especially infects domestic and wild cattle, and other ruminants may be similarly infected (ANONYM., 2004). In previous studies, cross-species infection bovine and caprine herpesviruses has been demonstrated (LEHMKUHL et al., 1985; YESILBAG et al., 2003). Detection of antibodies against BHV-1 in sheep suggest that sheep may play a role in the epidemiology of BHV-1 (ELAZHARY et al., 1984; JETTEUR et al., 1990; SURESH and SURIBABU, 1993), but sheep do not play a major role in BHV-1 transmission (HAGE et al., 1997).

The aim of this study was to investigate the prevalence of BHV-1 in sheep in the northern provinces of Turkey within a broad spectrum for the first time.

Materials and methods

Sheep blood samples (a total of 1146) were collected from animals of both sexes in eight locations in the northern provinces of Turkey and were shipped to our laboratory at 4 °C (Fig. 1). All sampled animals were unvaccinated.



Fig. 1. Map of Turkey showing the eight locations in the northern provinces of Turkey where serum samples were collected from sheep

Each blood sample was centrifuged at 3000 rpm for 20 min. at 4° C and sera were obtained. After centrifugation, each serum was poured into a separate tube (Eppendorf, Germany), heat inactivated at 56 °C for 30 min. and stored at -20 °C until tested.

Madin Darby Bovine Kidney cells (MDBK) were used for propagation, titrate and serum neutralization test for BHV-1. Cells were maintained at 37 °C in Dulbecco's minimal essential medium (DMEM, PAA, Inc., Austria) containing 10% heat-inactivated foetal bovine sera (FCS, PAA Inc., Austria) and DMEM containing 1% foetal serum. In the microneutralization test, DMEM was used with 10% foetal calf serum.

BHV-1 (Cooper strain, ATCC Cat. No. VR-864) was used in this study. The infectivity (TCID₅₀) of the virus was determined by the microtitre technique on MDBK cells as described by KAERBER (1964).

For investigation of antibody profile against BHV-1, the microneutralization method protocol was used, as previously described by FREY and LIESS (1971). Antibody titer at ½ or greater were accepted as positive.

Results

Infectivity of BHV-1 was calculated as 10^{7.0} TCID₅₀ /0.1 mL. The results of antibody profile against BHV-1 from sheep are detailed in Table 1. Overall percentage of BHV-1 in sheep tested was 1.74% (20/1146). Seropositivity rates were 0.51% (1/196), 4.5% (13/288), 2.4% (5/206), 0.58% (1/172) for the locations of Samsun, Tokat, Amasya and Sinop, respectively. All serum samples collected from sheep were negative for the locations of Ordu, Giresun, Rize and Trabzon.

Table 1. Seroprevalence of neutralizing antibodies against BHV-1 in sheep sera in eight different locations in northern Turkey

Province	N° of sheep tested	N° of positives (%)
Samsun	196	1 (0.51)
Tokat	288	13 (4.5)
Amasya	206	5 (5.2)
Sinop	172	1 (0.58)
Ordu	31	-
Rize	49	-
Giresun	99	-
Trabzon	105	-
Total	1146	20 (1.74)

The different age groups of tested animal are detailed in Table 2. Four hundred and eighty of 1146 sheep were aged between 14 and 24 months, and 666 sheep were aged above 24 months. Zero point eight three per cent (4/480) of sheep aged between 14 and 24 months age were found seropositive, while 2.40% (16/666) sheep aged above 24 months were found seropositive for BHV-1 antibodies.

Table 2. Distribution of antibodies against BHV-1 in the different age groups of the tested sheep

Age	N° of tested sheep	BHV-1 positive
Between 1 to 2 years	480	4 (0.83%)
Above 2 years	666	16 (2.40%)
Total	1146	20 (1.74)

The antibody titer for BHV-1 ranged between 1/2 and 1/18. Two of twenty positive serum (10%) had titres that ranged between 1/2 to 1/8, and 6 samples (30%) had titres that ranged between 1/32 to 1/64. Twelve (60%) had an antibody titer that ranged between 1/64 to >1/128.

Discussion

BHV-1 infects both domestic and wild cattle. Other ruminant species may also be infected with BHV-1 but no other reservoir of BHV-1, apart from ruminants, are known (ANONYM., 2004).

Through this, the first wide-ranging study in Turkey, the sero prevalence of BHV-1 in sheep was investigated. As a result, 20 of 1146 (1.74%) sheep were found to be seropositive. The results obtained from this study indicate that BHV-1 infection in sheep exists at low levels in Turkey. In the previous studies, serological evidence of BHV-1 in sheep had been similarly indicated. LEHMKUHL et al. (1985) reported that the prevalence of BHV-1 antibodies among lambs was between 0.4 and 5.4%. ELAZHARY et al. (1984) reported 10.8% BHV-1 seropositivity in tested sheep in Quebec, Canada, while JETTEUR et al. (1990) and CELEDON et al. (2001) detected that 8% tested sera were positive in Zaire and Chile. Prevalence of BHV-1 seropositivity in our study was determined to be between 0.51% and 5.2% depending on the province.

Reasons for this seropositivity obtained may be explained by the results of an experimental study by HAGE et al. (1997). In that study it was observed that sheep did not become easily infected with BHV-1 infection, and sheep which possess a latent infection with BHV-1 transferred the agent to cattle during reinfections at a minimal level. In seroprevalence studies performed by ALKAN et al. (1997; 2005) in Turkey, positivity

between 48.3 and 59.7% was determined in the fields we investigated. When the previous studies are compared to our present study, it is evident that BHV-1 infection is encountered rarely in sheep in the area, the reason for this being the restricted behaviour of the BHV-1 host range and the prevention by species barriers.

In conclusion, BHV-1 presence in sheep in the northern region of Turkey was determined, although no widespread presence was encountered.

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SAŽETAK

Uzorci seruma 1146 ovaca s osam različitih mjesta u sjevernom području Turske bili su pretraženi na govedi herpesvirus 1 (GHV-1) uobičajenim serum neutralizacijskim testom. Specifična protutijela za GHV-1 dokazana su u 1,74% (20/1146) ovaca. Protutijela za GHV-1 nisu bila dokazana u uzorcima uzetima od ovaca u četirima područjima. Time je prvi put dokazana zaraza tim virusom u ovaca u sjevernim provincijama Turske.

Ključne riječi: ovce, govedi herpesvirus 1, neutralizacijski test, seroprevalencija, Turska
