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RESEARCH ARTICLE

A serological investigation of Blue Tongue Virus infection in sheep breeds in Karaman province

Sibel Yavru[1], Oğuzhan Avcı[1], Orhan Yapıcı[1,2], Oya Bulut[1], Atilla Şimsek[1], Mehmet Kale[3]

[1]Department of Virology, Faculty of Veterinary Medicine, University of Selcuk, Konya, [3]Department of Virology, Faculty of Veterinary Medicine, University of Mehmet Akif Ersoy, Burdur, Turkey, [2]Faculty of Veterinary Medicine, University of Kyrgyzstan-Turkey Manas, Bishkek, Kyrgyzstan
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*oavci@selcuk.edu.tr

Karaman ilinde yetiştirilen koyunlarda Blue Tongue Virus enfeksiyonunun serolojik olarak araştırılması

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Öz

Amaç: Bu çalışma Karaman'da bulunan koyun işletmelerinde Blue Tongue Virus'a karşı seroprevalansın belirlenmesi amacı ile yapıldı.

Gereç ve Yöntem: Beş farklı işletmeden rastgele seçilen (her birinden 70 adet) toplam 350 koyundan kan serum örnekleri toplandı. Örnekler Blue Tongue Virus'a karşı gelişen antikor varlığı yönünden ticari olarak temin edilen competitive enzyme linked immunosorbent assay (cELISA) ile test edildi.

Bulgular: İşletmelerde Blue Tongue Virus'a karşı gelişen antikor prevalansı sırası ile %32.85, %28.57, %25.71, %37.14 ve %41.42 olarak belirlendi. Toplam 350 serum örneğinin 116 (%33.14)'sı Blue Tongue Virus'a spesifik antikor varlığı yönünden cELISA ile pozitif tespit edildi.

Öneri: Türkiye'nin iklim şartları Blue Tongue Virus'un vek-tör *Culicoides* türlerinin yaşamları için uygun olduğundan, koyunlar Blue Tongue Virus yönünden sürekli kontrol edilmelidir.

Anahtar kelimeler: Blue Tongue Virus, koyun, cELISA, *Cu-licoides.*

Abstract

Aim: The aim of this study was to describe the seroprevalence rate of Blue Tongue Virus in sheep flocks in Karaman.

Materials and Methods: A total of 350 sheep blood serum samples were collected from 5 flocks (70 from each flocks) that were randomly selected. Samples were tested against to Blue Tongue Virus antibodies by a commercial competitive enzyme linked immunosorbent assay (cELISA).

Results: Prevalence of antibodies to Blue Tongue Virus in flocks was 32.85%, 28.57%, 25.71%, 37.14%, 41.42%, respectively. Out of 350 serum samples, 116 (33.14%) were positive for Blue Tongue Virus specific antibodies by cELISA.

Conclusion: The climate conditions of Turkey might be suitable for the survival of *Culicoides* vectors of Blue Tongue Virus; hence sheep folks should be controlled constantly in term of BTV.

Keywords: Blue Tongue Virus, sheep, cELISA, *Culicoides.*

Introduction

Bluetongue (BT), Office International Epizooties (OIE) lists A viral disease, is a vector-borne disease in domestic and wild ruminants caused by Orbivirus genus of the family Reoviridae (Attoui et al 2009, Matsuo and Roy 2013). It can be generally transmitted by biting midges of the genus Culicoides (Bishop et al 2000, Nayduch et al 2014). Twenty six distinct serotypes have been reported (Maan et al 2011, 2014). BTV is an arthropod-borne (Roy and Noad 2006) orbivirus that causes important viral disease mainly in sheep and less frequently in cattle, goats, deer, elk, camels, and wild ruminants (Savini et al 2007, Mellor et al 2008, Arenas-Montes et al 2014). BTV can play an important role as a viral pathogen in abortive cases in sheep (Zientara and Ponsart 2014).

Different serological diagnostic methods (Agar gel immunodiffusion, hemagglutination inhibition, and competitive-ELISA) have been used to detect serogroup of BTV (Ward et al 1995, Kramps et al 2008, Mozaffari et al 2014). It is known that there is more immunological cross-reactivity among BTV serogroups (Maclachlan et al 2014). Although BTV serotypes are differentiated on the basis of genotype; neutralization test (Patton et al 1994, Maan et al 2014) can be used for detection of serotype of BTV. Reverse transcription polymerase chain reaction (RT-PCR) can be used for direct detection of BTV in blood or tissue samples (Aradaib et al 2003, 2005, Maan et al 2012).

It has been hypothesized that BTV infection which reported from different region of central Anatolia may affect sheep in Karaman. The aim of this study was to determine the first data status of BTV infection in Karaman.

Material and Methods

Totally five different flocks (70 sheep in each flocks) all sampled sheep (totally 350; randomly selected in June, July and August of 2010 in Karaman) were unvaccinated for BTV. All applications were conducted according to the animal welfare. All sampled animals were Merinos and female. Blood samples were packed in dry ice and were brought Virology Laboratory, Faculty of Veterinary Medicine, University of Selcuk and centrifuged at 3000 × g for 10 min for serum preparation. Approximately 1 mL of serum was collected into sterile eppendorf tubes and stored at -20°C until analysis. Sera were tested for antibodies against to BTV by a commercially available competitive ELISA (cELISA, Veterinary Medical Research and Development Inc., Pullman, WA, USA). The test was performed according to manufacturer's instructions. The optical densities of plates were read with an automatic ELISA plate reader (Rayto RT-2100C, China). The percent inhibition (%) values of positive, negative controls and samples were calculated. Statistical significance of differences between provinces was calculated by using chi-square test (Minitab 14.0 Inc., State College, PA, USA). Difference were considered significant when $P<0.05$.

Table 1. Seroprevalence of BTV in flocks.

Number of flock	Number of samples	BTV Antibody (+)	Seroprevalence rate (%)
1	70	23[a]	32.85
2	70	20[a]	28.57
3	70	18[b]	25.71
4	70	26[a]	37.14
5	70	29[a]	41.42
Total	350	116	33.14

[a, b:] Different letters in same column are statistically significant ($P<0.05$)

Results

Seroprevalence rates of specific antibodies against BTV are shown in Table 1. Totally seroprevalence rate of BTV was determined as 33.14%.

Discussion

Bluetongue is an important and abortive infection of both domestic (Sheep, goat, camels, etc) and wild ruminants which clinical characterized by different symptoms such as congestion, cyanosis of the tongue, hemorrhage near the base of the pulmonary artery, oedema, reduced wool quality, poor subsequent reproductive performance, decrease milk production (Verwoerd and Erasmus 2004, Aradaib et al 2005, Gür 2008, Maclachlan et al 2008, 2009). BTV infection is a seasonal infection (Carpenter et al 2013) and may cause economic losses (van der Sluijs et al 2012) in flocks and transmitted by Culicoides species; therefore, the prevalence of the BTV infection increases during the spring, summer, and fall when the density of Culicoides increases (Tabachnick 1996, Meiswinkel et al 2008, Mellor et al 2008, Darpel et al 2011, van der Sluijs et al 2012).

In this study, 33.14% seropositivity for BTV was detected by cELISA, widely used for rapid and serological diagnosis of BTV infection of flocks, in unvaccinated sheep in Karaman (Table 1). It has been reported that climate is a major risk factor for BTV infection (Purse et al 2005, 2008, Guis et al 2012). All flocks were determined as seropositive for specific antibodies for BTV. Out of 350 serum samples, 116 were positive (Table 1). Prevalence of antibodies to BTV in flocks was 32.85%, 28.57%, 25.71%, 37.14%, 41.42%, respectively. In five seropositive flocks the prevalence ranged from 25.71% to 41.42%. There was no statistically difference about serological detection of BTV infection between 1, 2, 4, and 5 while 3th flocks were significantly low results (Table 1). Seropositive results obtained from this study could depend on sampling time and humid climate but unfortunately there is

no consideration about possible vector specious. In addition seropositive results indicates natural infection in sheep because lack of as a vaccination programme for BTV infection in Karaman. BTV infection was reported by different researchers (Yavru et al 1997, Bulut et al 2006) in central region of Anatolia.

Results obtained from this study demonstrate that BTV infection affects sheep in Karaman located in Central region of Anatolia. BTV infection can lead economics losses due to defect on animals and decreased of animal products (Velthuis et al 2010) and should be limited transportation of animals from a point to another (Maclachlan and Osburn 2006). It can be useful that control, prevention and vaccine programmes of BTV infection in region due to transmission by Culicoides (Maclachlan and Mayo 2013).

Conclusion

This is the first knowledge of sheep infected by BTV in Karaman. Further studies on various animal species potential vectors are needs to establish possible vector species, the serological and virological evidence of BTV serotypes that are circulating in central Anatolia including Karaman.

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