



Investigation of Bovine Viral Diarrhoea Virus in Dairy Cattle Premises Where Abortions Occur*

Metin GÜRÇAY¹, Mustafa İSSİ², Yusuf GÜL²

¹ Republic of Turkey, Ministry of Food Agriculture and Livestock, Veterinary Control and Research Institute, Elazığ-TURKEY

² University of Firat, Faculty of Veterinary Medicine, Department of Internal Disease, Elazığ-TURKEY

Summary: In this study, the presence of Bovine Viral Diarrhoea Virus (BVDV) was investigated by serologic and virologic methods in dairy cattle enterprises where abortions occur. Study materials include 602 blood samples taken from aborted cows and 241 tissue samples taken from waste fetus samples (abortion/stillborn fetus) admitted to the Control Institute of Veterinary Medicine (Elazığ) from dairy cattle enterprises where abort cases were seen in Elazığ, Malatya and Bingöl between September 2011 and April 2012. The lung, liver, spleen and brain tissue samples taken from aborted fetus and blood serum samples were tested by ELISA. With regard to BVDV antibodies, the serum blood samples were found to be positive in 70.76% (426/602) in total. This ratio was determined as 76.11% (137/180), 56.25% (18/32), 69.48% (271/390) in Elazığ, Malatya and Bingöl provinces, respectively. The prevalence of BVDV antigen in the tissue samples of aborted fetuses was detected as 28.63% (69/241) in total, 25.75% (17/66) in Elazığ, 36.36% (4/11) in Malatya and 29.26% (48/164) in Bingöl.

As a result, in aborted dairy cows in Elazığ, Malatya and Bingöl, antigens and antibodies have been reported to high-seroprevalance. It was noted that in order to reduce economic losses due to this virus, there should be emphasis on preventive measures.

Key Words: Abortion, BVDV, dairy cattle, ELISA

Abort Olgularının Görüldüğü Süt Sığırı İşletmelerinde Bovine Viral Diarrhoea Virusun Araştırılması

Özet: Bu çalışmada, yavru atma olaylarının görüldüğü süt sığırı işletmelerinde Bovine Viral Diarrhoea Virus (BVDV)'un varlığı serolojik ve virolojik olarak araştırıldı. Çalışma materyalini, Eylül 2011 ile Nisan 2012 ayları arasında Elazığ, Malatya ve Bingöl illerinde yavru atma olaylarının görüldüğü süt sığırı işletmelerinden Elazığ Veteriner Kontrol Enstitüsüne gönderilen 602 adet inek kan serumu ile 241 adet atık numunesinden (abort/ölü doğmuş fötüsten) alınan doku örnekleri oluşturdu. Fötüse ait akciğer, karaciğer, dalak ve beyin doku örneklerinden hazırlanan doku ekstraktları ile kan serumu örnekleri ELISA testi ile incelendi. Kan serumu örneklerinde BVDV antikorları yönünden genel toplamda %70.76 (426/602) belirlenirken Elazığ ilinde %76.11 (137/180), Malatya ilinde %56.25 (18/32) ve Bingöl ilinde %69.48 (271/390) oranında pozitiflik tespit edildi. Atık numune örneklerinin ise toplamda ve illerde sırasıyla %28.63 (69/241), %25.75 (17/66), %36.36 (4/11) ve %29.26 (48/164) oranında BVDV antijenleri yönünden pozitif olduğu belirlendi. Sonuç olarak Elazığ, Malatya ve Bingöl bölgesinde atık yapmış süt ineklerinde BVDV antijen ve antikorlarının yüksek seroprevalansa sahip olduğu ve bu virusa bağlı ekonomik kayıpların önlenmesi için koruyucu tedbirlere önem verilmesi gerektiği düşünülmektedir.

Anahtar Kelimeler: BVDV, ELISA, süt sığırı, yavru atma

Introduction

Included in pestivirus genus of Flaviridae family, Bovine Viral Diarrhoea Virus (BVDV) infections are observed worldwide as well as in our country and cause significant economic losses (1, 18, 20). Virus with genotype of BVDV-1 and BVDV-2 has two

biotypes according to their cytopathic effects on cell cultures namely; cytopathogenic (cp-BVDV) and noncytopathogenic (ncp-BVDV) biotypes (16, 18, 20).

In cattle with BVDV, bovine viral diarrhoea, acute or chronic mucosal disease, congenital anomalies or persistent infected calves (virus emitting), abortion and calves with low chance of survival due to transplacental infections have been reported in various clinical manifestations (1, 18).

In this study, we intend to analyze abortions caused by BVDV infections with serological and

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virological aspects, in dairy cow enterprises in Elazığ, Malatya and Bingöl.

Material and Methods

Study materials include tissue samples taken from 241 waste fetus samples (abortion/stillborn fetus) seen in the events of abortion and sent to the Control Institute of Veterinary Medicine (Elazığ) for etiological diagnosis at small family-type dairy cow enterprises and 602 units of blood samples taken from clinically healthy aborted cows in these enterprises between September 2011 and April 2012 in Elazığ, Malatya and Bingöl (Table 1). Anamnesis revealed that none of these cows received protective vaccination for BVDV and calves born had no chance of living between 6 and 9 months of pregnancy.

BVDV Antigen Test from Tissues of Aborted Fetuses

In order to research BVDV antigen, brain, liver, lung and spleen tissue samples taken from aborted fetus were stirred with 2 ml PBS diluents (1/10). Also MagNaLyser (Roche, Mannheim, Germany) were homogenized in beaded tubes for 3000 xg and 3 minutes. Homogeneta Eppendorf tubes were centrifuged for 10 minutes and 3000 xg and solid materials and beads were removed. Supernatant that remained solution was stored at -80 °C until used as inoculums. The presence of antigens in inoculums was investigated with commercial BVDV Ag ELISA kit (BVDV Ag Serum Plus HerdChek IDEXX Laboratories Westbrook, Maine 04 092 USA). Results were analyzed at 450 nm absorbance filter and as sample-negative control were >0.300 nm, BVDV antigen was evaluated as positive (7, 14, 24).

Table 1. Distribution by province of the number of samples obtained from waste fetus samples taken from dairy cow enterprises and blood serum samples

City	Fetus tissue samples		Blood serum samples	
	n	%	n	%
Elazığ	66	27.38	180	29.90
Malatya	11	4.56	32	5.31
Bingöl	164	68.04	390	64.78
TOTAL	241		602	

Table 2. Results of BVDV antigen and antibody screening test of dairy cows included in study

City	BVDV antigen ELISA results of fetus tissue samples					BVDV antibody ELISA results of blood serum samples				
	n	-	%	+	%	n	-	%	+	%
Elazığ	66	49	74.24	17	25.75	180	43	23.88	137	76.11
Malatya	11	7	63.63	4	36.36	32	14	43.75	18	56.25
Bingöl	164	116	70.73	48	29.26	390	119	30.51	271	69.48
TOTAL	241	172	71.36	69	28.63	602	176	29.23	426	70.76

Obtaining of Blood Sera and BVDV Antibody Test

For BVDV antibody search, blood taken from *v. jugularis* of cows was centrifuged at 3000 xg for 10 minutes. Serum remained above was transferred into sterile tubes and stored at -20 °C until use. Presence of antigens in blood was investigated with commercial BVDV total antibody ELISA test kit (IDEXX Laboratories Westbrook, Maine 04092 USA) (8).

Statistic Analysis

For analyzing the difference between the prevalence of the diseases amongst the provinces, Chi-Square test form SPSS software program (Version 13.0) was used (10).

Results

Results of BVDV antigen and antibody screening tests of fetal tissue and blood serum samples of aborted dairy cows are shown at Table 2. It was determined that the difference between the provinces regarding BVDV antigens of the fetal tissues was not significant ($P>0.05$) whereas that between the same regions in relation to BVDV antibodies was significant ($P<0.05$) among Elazığ-Malatya and Malatya-Bingöl (Table 3).

Discussion

In postnatal infections due to BVDV commonly seen in ruminants, fertility disorders can be seen. Also in transparental infections, abortions depending on stage of fetal development, embryonic death and resorption of fetus, congenital anomalies, birth of weak calves and newborn calves with appearance healthy but

seropositive immune tolerance and persistent infection have been reported. Reports show that calves with persistent infections play an important role at BVDV contamination. Although they seem healthy, they spread virus with their secretions and excretion and this factor plays an important role in scattering of the disease to other animals (1, 18). In addition, reports (1, 18, 20) indicate that deaths due to gastrointestinal and respiratory tract diseases cause large economic losses.

According to Ok (16) abortions caused by BVDV infections can be seen any time at pregnancy and cases of abortion are observed in animals at a variety of periods of pregnancy.

BVDV infections are common worldwide (18, 20) and there have been studies reporting the presence of BVDV infections in our country (5, 11, 17, 23, 25) and in our region (9). Bovine Viral Diarrhoea Virus infections have been reported to have very different seroprevalence (2-6, 11-13, 15, 17, 19, 21-23, 25). Seropositivity has been reported as 58-93% in South Vietnam (6), 5% Switzerland (19), 21% in Spain (13), 35% in North Portugal (15), 84.4% in Britain (2), 32% in Sweden (3), 38% in South Korea (12), 14% in Mexico (21), 94% in Peru (22), 79.2% in Croatia (4) and in our country 86% in Aydın vicinity (23), 81.62% in Northeast Anatolia region (25), 90.63% in Konya region (11) and 82.1%, 39.49%, 83.4% and 78.6% with a total of 64.2% in Ankara, Konya, Muğla ve Samsun respectively (5). Also, in Afyonkarahisar, seropositivity has been found to range between 77.7% and 100% in organized flocks and 57.1% in family-type farms (23).

In this study, in terms of BVDV antibodies in blood sera, results were found to be positive in Elazığ as 76.11% (137/180), in Malatya 56.25% (18/32) and in Bingöl 69.48% (271/390), according to waste

Table 3. The evaluation significance of the diseases between the provinces

	Fetus tissue samples		Blood serum samples	
	χ^2 value	Statistical significant	χ^2 value	Statistical significant
Elazığ-Malatya	0.134	$P>0.05$	4.489	$P<0.05$
Elazığ-Bingöl	0.189	$P>0.05$	2.34	$P>0.05$
Malatya-Bingöl	0.329	$P>0.05$	20.59	$P<0.001$

samples, seropositivity for BVDV antigens was found as 25.75% (17/66), 36.36% (4/11) and 29.26% (48/164) in Elazığ, Malatya and Bingöl, respectively.

For the determining presence of virus at fetal tissues methods such as virus isolation, RT-PCR and ELISA are used (7, 8, 14, 24). In a comparative study (7), diagnosis of BVDV in blood and fetal fluids were determined with RT-PCR and Ag ELISA, results of both tests are reported as similar. Same researchers stated that Ag ELISA a valuable test method for the diagnosis of BVDV at routine laboratories owing to its good performance, low price, and it can even be used on autolyzed fetuses. In this study, BVDV antigen and antibodies in blood serum and fetal tissue samples were screened with ELISA test kits.

Due to lack of treatment options, in order to prevent BVDV infections, first of all animals with persistent infection should be removed from herd and vaccination programs should be set (1, 16, 18, 20).

As a result, in aborted dairy cows in Elazığ, Malatya and Bingöl, antigens and antibodies have been reported to high-seroprevalance. It was noted that in order to reduce economic losses due to this virus, there should be emphasis on preventive measures.

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Correspondence Address :

Doç. Dr. Mustafa İSSİ
Fırat Üniversitesi Veteriner Fakültesi,
İç Hastalıkları Anabilim Dalı,
23119, Elazığ, TÜRKİYE
Tel: +90424 2370000 / 3874
E-posta: mustafaissi@hotmail.com