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Short communication

Prevalence of Persistent Bovine Viral Diarrhea Infection in Industrial Dairies of Tehran and Alborz Province, Iran

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Abstract

Background and Aims: Bovine viral diarrhea virus (BVDV) is considered as worldwide and economically significant infection. The aim of this study was to estimate prevalence of persistent bovine viral diarrhea infection (PI) in industrial dairies of Tehran and Alborz province, Iran.

Materials and Methods: In this cross-sectional study, 102371 blood samples were collected from cattles of industrial dairies of Tehran and Alborz province during 2018. Indirect enzyme-linked immunosorbent assay (ELISA) and Reverse transcription-polymerase chain reaction (RT-PCR) were performed for determination of viral genotypes, including BVDV1, BVDV2, and HOBi like among PI samples.

Results: A total of 102371 samples were collected from 76 industrial dairies in Tehran and Alborz province, that 512 cases were PI. All samples were positive for BVDV1 (100%). Other genotypes were not found among PI samples.

Conclusion: Prevalence of persistent bovine viral diarrhea infection in industrial dairies in Tehran and Alborz was not high. However, it will be further decreased by implementing control and eradication programs.

Keywords: Bovine Diarrhea Virus; Dairy industry; Prevalence

Introduction

Bovine viral diarrhea virus (BVDV) is an RNA virus belonging to Pestivirus genus, that is classified in Flaviviridae family (1). Two genotypes of BVDV (BVDV-1 and BVDV-2) are characterized based on the nucleotide sequence of the 5'untranslated region (5'-UTR) of viral genome (2). In addition, Hobi-like virus- an emerging pestivirus- is introduced as BVDV-3. BVDV-1 currently has been classified into 21 subtypes (1a−1u) and BVDV-2 into four subtypes (2a−2d) (3).

Two biotypes of BVDV are identified based on the cytopathic effect of virus in the cell culture system. A cytopathic (CP) biotype induces apoptosis and death of target cells, while a noncytopathic (NCP) biotype replicates in cell culture without damage to infected cells. The NCP biotypes are the most abundant isolates (about 95%) in nature (4).

BVDV is transmitted among susceptible animals through direct contact, venereal transmission or vertical exposure (5). Respiratory disease, reproductive failure, congenital defects, mucosal disease, diarrhea and thrombocytopenic (bleeder) syndrome are the common symptoms of the infection; however, the disease occurs in most animals without clinical signs (6). BVDV infection can affect the pregnant animals and lead to teratogenesis, fetal abortion, embryonic resorption, mummi-

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fication or stillbirth. BVDV persistent infection (PI) occurs when the virus infects the fetus during the first trimester of gestation and results in a continuing reservoir of infection.
Transient or persistent infection with BVDV causes immunosuppression, which predisposes animals to other infections (7).
BVDV infection is an economic concern in cattle industry. This infection has been reported from different regions of Iran (8, 9).
In recent years, use of inactivated vaccines in some industrial dairy farms has been decreased significant economic losses caused by BVDV. This study aimed to obtain more epidemiological information about the disease in Tehran and Alborz province, Iran.

Methods

Sampling: In this cross-sectional study, 102371 blood samples were collected from cattles of industrial dairies of Tehran and Alborz province during 2018. Two samples from each cow were taken at two weeks intervals. Samples were centrifuged (10 min at 3000 rpm) to separate sera, then transported on ice to laboratory and stored at - 20 °C until processing.
ELISA test: Serum samples were evaluated for BVDV antibodies using a commercial ELISA kit (IDEXX BVDV, Switzerland, Liebefeld-Bern) according to the manufacturer's procedures. This kit has 95% sensitivity and 98% specificity. The optical density (OD) was measured at 450 nm .
RNA extraction and RT-PCR: viral RNA was isolated from 100 µl sera using High Pure RNA Isolation Kit (Roche Applied Sciences, Germany) following the manufacturer's procedure. Extracted RNA was converted to cDNA using the RevertAid First Strand cDNA synthesis kit (Thermo Scientific; USA). The RT-PCR reaction was performed in a total volume of 25, containing 12.5µl master mix, 0.5µl of each specific primer, including forward: 5′- GCCATGCCCTTAGTAGGACT-3, reverse for BVDV1: 5′- GCAGCACCCTA TCAGGCTGT-3′, reverse for BVDV2: AGA TCG GTC CTGGTT TGA TA, revere for HOBi like: 5′-TCGACGCATCAAGGAATGC CT-3′, 6.5 µl DEPC, and 5 µl of template. The PCR was performed using thermal cycling as follows: one cycle at 94 °C for 5 min followed by 35 cycles in 3 continuous phases including 94 °C for 30s, 50 °C for 30s, and 72 °C for 30s and finally one cycle at 72 °C for 10min. The PCR products were separated by agarose gel electro-phoresis (1.5%), stained with DNA safe stain, and examined under UV transluminator. A positive control for RNA-BVDV and a negative control were included in each amplification run. Amplification products of 235, 546 and 655 bp were predicted for BVDV type I, II and III respectively.

Results

A total of 102371 samples were collected from 76 industrial dairies of Tehran and Alborz province, that 512 cases were seropositive and classified as PI. Fifty samples diagnosed as PI were genotyped by PCR method for determination of viral genotypes, including BVD1, BVD2, and HOBi like. All 50 samples had the genotype BVDV1 (Fig. 1). Other genotypes were not found among PI samples.

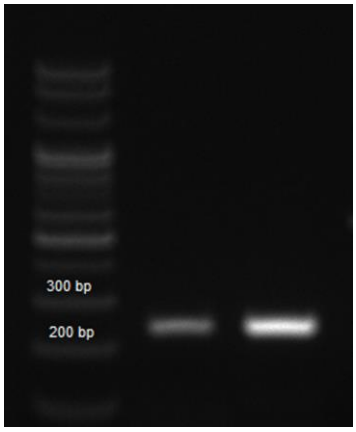

Fig. 1. Electrophoresis of PCR products of BVDV1 genome. The amplicon size was 235 bp.

Discussion

Several factors can cause abortion in livestock, but the causes of animal abortion are not clear in more than 50% of the cases. BVD is an important viral disease which causes infertility and abortion in animals. BVDV can induce changes in fetal placenta barrier that allow

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other pathogenic organisms to cross it (10). The prevalence of persistently infected (PI) cattle has typically been reported in the range of 0.5% to 2%. Although the prevalence of BVDV PI can be high (25-30%) when a large number of naïve animals, early in pregnancy, have been exposed to noncytopathic BVDV (11).

In the present study, the prevalence of persistent BVD infection in industrial dairies of Tehran and Alborz province was 0.5%.

In consistent with our finding, Erfani et al. were reported that the prevalence of BVDV PI was 0.53% in Zanjan province (1). The reported rate of PIs in some studies conducted in Iran was higher than the current study. The prevalence of PI in a study performed in Fars province was 4% (12). In study of Garoussi et al., the rate of BVD persistent infection among dairy cattle herds in the suburb of Mashhad was 1.42% (13). In 2001, Rohani et al reported that the 7.4% of calves in dairy farms of Tehran area persistently infected with BVD virus (14). They reported a higher rate of PI infection among calves in comparison with our results. The observed difference may be due to differences in the laboratory methods (ELISA kit) or sample sizes. They studied 375 samples, but the current study performed on 102371 samples. In addition, the present study is conducted in 2018 when the control programs were more developed than in previous years. The effective prevention and control strategies have a significant role in decreasing of PI BVDV. Performing nationwide bovine viral diarrhea control for Six years (2011–2016) in Germany had decreased PI cases from 3.44% to 0.16% (15).

Genotype BVDV-1 is predominant in most part of the world, whereas BVDV-2 represents the majority of cases in North America. In Europe, BVDV-2 was first detected in the United Kingdom in 2000 and currently causes up to 11% of BVD cases (16). In the current study, only genotype BVDV1 was identified and other genotypes were not founded. Fulton et al. previously obtained same data in USA.

They reported 25 PI calves from 4530 samples (0.55%) and genotype of all the PI isolates were BVDV1 (17). In a study conducted in Colombia BVDV1 was identified as a single genotype for all the samples (18). In the study of Khodakaram-Tafti et al. in Fars province, BVDV1 was the predominant genotype (75%) followed by BVDV2 (25%) (19). Similar to our finding, Mosakhani et al. were found BVDV1 in 100% of PI-BVDV samples collected from industrial dairies of Tehran. The sample size in their study (20 samples) was less than our study, but they detected BVDV2 in 3 samples (20). A new type of BVDV called HoBi-like (BVDV3), have been identified in Brazil, Southeast Asia, and Europe (16). We did not detect BVDV3 in our samples. This genotype has not been reported in any study from Iran.

Conclusion

Prevalence of persistent bovine viral diarrhea infection in industrial dairies of Tehran and Alborz was not high. However, it will be decreased by developing control and eradication programs.

Acknowledgment

Not applicable.

Conflict of interest

The authors declare no conflict of interest.

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