

< Short Communication >

## Serological and virological investigation of pestiviruses in Korean black goats

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### Abstract

Blood samples were collected from 672 goats in 60 farms from five provinces of Korea between November 2009 and August 2011. The prevalence of antibodies to pestiviruses was investigated. The examination for antibodies was performed using an enzyme-linked immunosorbent assay (ELISA) detecting antibodies to the bovine viral diarrhea virus (BVDV) and border disease virus (BDV). All blood samples were screened using reverse transcription-polymerase chain reaction (RT-PCR) with primer pairs specific to common pestivirus genome regions. The observed individual seroprevalence was 1.49% and herd seroprevalence was 11.67%. Also, the specific genomes to pestiviruses were detected in 3 out of the 915 clinical samples (0.45%). Based on the nucleotide sequence data, detected pestiviruses were belonged to two BVDV type-1 and one BVDV type-2. The pestivirus infection has been occurred among Korean black goats. However, our results indicate that the prevalence of pestiviruses in black goats was not significantly higher on farms with cattle.

**Key words :** Prevalence, Pestiviruses, BVDV, Black goat

### INTRODUCTION

The genus *Pestivirus* within the family *Flaviviridae* includes *Bovine viral diarrhea virus* (BVDV), *Border disease virus* (BDV), *Classical swine fever virus* (CSFV) and a tentative 'Giraffe' species (Fauquet et al, 2005). Recently, atypical pestivirus species, proposed as BVDV-3, was also isolated from clinically affected calves in Italy (Decaro et al, 2011). BDV infection is mainly found in sheep, causing abortion, stillbirths, and /or weak offspring and BVDV infections are endemic in cattle worldwide (Houe, 1995; Løken, 1995). Infection with pestiviruses can result in severe economic and reproductive losses. Pestiviruses can cross the host species barrier and infected a wide range of host (Nettleton and

Entrican, 1995). Under field conditions, the transmission of BDV from ruminants to cattle is relatively uncommon. However, the transmission of BVDV from cattle to sheep and goat vice versa has been described previously (Paton et al, 1997).

Serological studies have shown a worldwide distribution of BDV in sheep and goat. A BD-like syndrome in goats has been reported in southern part of Korea since 1998. In a retrospective study, BVDV-2 in native goat was identified and characterized in 2005 (Kim et al, 2006). BDV have not been identified in Korean goats until now.

In Korea, several investigations of the prevalence and characterizations of BVDV have been performed in cattle (Lee et al, 2008; Oem et al, 2010; Oem et al, 2009; Park et al, 2004). However, the survey of infectious diseases in goats has rarely performed. In particular, a few

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studies of prevalence of pestivirus in Korean goats have been performed (Yang et al, 2008). The Korean black goat population in Korea numbers about 266,000 animals distributed in 21,000 herds. The population of goats in Korea is increasing every year. The aims of the present study are to investigate the prevalence of pestiviruses in Korean black goats and determine the predominant pestiviruses in the goat populations.

## MATERIALS AND METHODS

### Animals and clinical samples

In this study, large-scale survey was conducted to investigate the infection of pestiviruses in Korean black goats. Only farms with 100 or more goat herds were included in the study. One each farm, approximately 10 goats were randomly selected and sampled. A total of 672 black goats were sampled from 60 farms of a variety of breeds. Clinical samples (672 whole bloods, 150 feces and 93 nasal swabs) were collected from November 2009 to August 2011. The all samples obtained were kept  $-70^{\circ}\text{C}$  until tested.

### ELISA and RT-PCR

An ELISA kit (Svanova Biotech, Sweden) was used to detect antibodies for BVDV/BDV as the manufacturer's instructions. Also, viral RNA was extracted from 915 clinical samples (672 whole bloods, 150 feces, 93 nasal swabs) using an RNeasy Mini Kit (Qiagen, Valencia, CA, USA). RT-PCR procedures were performed in a one-tube system using the pan-pestivirus primer pair V324/V326; this system amplifies a portion of the 5'-UTR of pestivirus genomes (Vilcek et al, 1994). The amplified 297-bp DNA fragment was purified using an Agarose Gel DNA Extraction Kit (INTRON, Daejeon, Korea) and subcloned into the vector pGEM-T (Promega, Madison, WI, USA) according to the manufacturer's instructions.

Automated nucleotide sequencing of the 5-UTR gene inserted into the vector was performed on an ABI 3130XL genetic analyzer (Applied Biosystems, Foster

City, CA, USA) with the Big Dye Terminator cycle sequencing kit (Applied Biosystems). All nucleotide positions were confirmed by three or more independent sequencing reactions in both directions.

## RESULTS AND DISCUSSION

Antibodies to pestiviruses were detected in 10 of the 672 serum samples (1.49%), and 7 of the 60 herds (11.67%) were seropositive for pestiviruses (Table 1). Also, the specific genomes to pestiviruses were detected in 3 of the 915 clinical samples (0.45%) and 3 of the 60 herds (5%) were detected for pestiviruses (Table 1). Based on the nucleotide sequence data, Sequence analysis of 5'-UTR region showed that two pestiviruses were typed as BVDV-1b and the other pestivirus belonged to BVDV type-2. No BDV was detected by RT-PCR and sequencing in this study (Data not shown).

The overall prevalence of pestivirus infection varies from 2 to 75.9% in sheep and goats depending the countries (Krametter-Frötscher et al, 2006; Krametter-Frötscher et al, 2007; Mishra et al, 2009; Tutuncu et al, 2011). The seroprevalence (1.49%) detected in this study is lower than previously reported studies in several countries (Krametter-Frötscher et al, 2006; Krametter-Frötscher et al, 2007; Mishra et al, 2009; Tutuncu et al, 2011). This situation could be explained with different factors. First, BDV infection in Korean goat population has not been reported until now. Secondly, a number of goat farms are not closely adjacent to other ruminant farms. Third, no vaccine has been used to prevent pestivirus infection in goat.

In this study, three pestiviruses were identified. Two

**Table 1.** Prevalence of pestiviruses in Korean black goats (*Capra hircus aegagrus*)

Items	Method tested	
	ELISA	RT-PCR
No. of herds tested	60	60
No. of goats tested	672	915
Herd prevalence (No. of positive)	11.67 (7)	5 (3)
Goat prevalence (No. of positive)	1.49 (10)	0.33 (3)*

\*All pestiviruses were detected in blood samples.

pestiviruses were typed as BVDV-1b and one pestivirus as BVDV-2. Interestingly, two BVDV-1b and one BVDV-2 were very closely related to KA07 strain and KA09 strain described previously in Korea, respectively (Oem et al, 2009). These results indicate that the identified three pestiviruses might be originated from cattle. Persistently infected (PI) goats are uncommon and it seems reasonable to assume that PI cattle are the main reservoirs for pestivirus infections in the goat (Løken, 1995). The prevalence of BVDV in dairy cows in Korea was investigated and 58% cattle were seropositive (Lee et al, 2008). It may be evidence that goat pestiviruses were transmitted from cattle. In Korea, vaccination to reduce the number of BVD outbreaks has been performed but compulsory notification and national eradication programs are not yet carried out. However, these programs need to reduce economic losses by BVDV outbreaks in near future in Korea. Our results in this study will benefit to perform these programs as supportive data.

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## REFERENCES

- Decaro N, Lucente MS, Mari V, Cirone F, Cordioli P, Camero M, Sciarretta R, Losurdo M, Lorusso E, Buonavoglia C. 2011. Atypical pestivirus and severe respiratory disease in calves, Europe. *Emerg Infect Dis* 17: 1549-1552.
- Fauquet CM, Mayo MA, Maniloff J, Desselberger U, Ball LA. 2005. Virus taxonomy, classification and nomenclature of viruses. Eighth report of the international committee on taxonomy of viruses, 1162. Elsevier Academic Press, London.
- Houe H. 1995. Epidemiology of bovine viral diarrhea virus. *Vet Clin North Am Food Anim Pract* 11: 521-547.
- Kim IJ, Hyun BH, Shin JH, Lee KK, Lee KW, Cho KO, Kang MI. 2006. Identification of bovine viral diarrhea virus type 2 I Korean native goat (*Capra hircus*). *Virus Res* 121: 103-106.
- Krametter-Fröetscher R, Loitsch A, Kohler H, Schleiner A, Schiefer P, Moestl K, Golja F, Baumgartner W. 2006. Prevalence of antibodies to pestiviruses in goat in Austria. *J Vet Med B Infect Dis Vet Public Health* 53: 48-50.
- Krametter-Fröetscher R, Loitsch A, Kohler H, Schleiner A, Schiefer P, Mostl K, Golja F, Baumgartner W. 2007. Serological survey for antibodies against pestiviruses in sheep in Austria. *Vet Rec* 160: 726-730.
- Lee DH, Park SW, Choi EW, Lee CW. 2008. Investigation of the prevalence of bovine viral diarrhea virus in dairy cows in South Korea. *Vet Rec* 162: 211-213.
- Løken T. 1995. Ruminant pestivirus infections in animals other than cattle and sheep. *Vet Clin North Am Food Anim Pract* 3: 597-614.
- Mishra N, Rajukumar K, Tiwari A, Nema RK, Behera SP, Satav JS, Dubey SC. 2009. Prevalence of bovine viral diarrhea virus (BVDV) antibodies among sheep and goats in India. *Trop Anim Health Prod* 41: 1231-1239.
- Nettleton PF, Entrican G. 1995. Ruminant pestiviruses. *Br Vet J* 151: 615-642.
- Oem JK, Hyun BH, Cha SH, Lee KK, Kim SH, Kim HR, Park CK, Joo YS. 2009. Phylogenetic analysis and characterization of Korean bovine viral diarrhea viruses. *Vet Microbiol* 139: 356-360.
- Oem JK, Chung JY, Roh IS, Kim HR, Bae YC, Lee KH, Jin YH, Lee OS. 2010. Characterization and phylogenetic analysis of bovine viral diarrhea virus in brain tissues from nonambulatory (downer) cattle in Korea. *J Vet Diagn Invest* 22: 518-523.
- Park JS, Moon HJ, Lee BC, Hwang WS, Yoo HS, Kim DY, Park BK. 2004. Comparative analysis on the 5'-untranslated region of bovine viral diarrhea virus isolated in Korea. *Res Vet Sci* 76: 157-163.
- Paton D, Gunn M, Sands J, Yapp F, Drew T, Vilcek S, Edwards S. 1997. Establishment of serial persistent infections with bovine viral diarrhea virus in cattle and sheep and changes in epitope expression related to host species. *Arch Virol* 142: 929-938.
- Tutuncu M, Duz E, Karaca M, Akkan HA, Keles I, Bakir B, Tasal I. 2011. A serological investigation of pestiviruses in sheep in eastern border of Turkey. *Trop Anim Health Prod* 43: 1467-1469.
- Vilcek S, Herring AJ, Herring JA, Nettleton PF, Lowings JP, Paton DJ. 1994. Pestiviruses isolated from pigs, cattle and sheep can be allocated into at least three genogroups using polymerase chain reaction and restriction endonuclease analysis. *Arch Virol* 136: 309-323.
- Yang DK, Kweon CH, Kim BH, Choi CU, Kang MI, Hyun BH, Hwang IJ, Lee CS, Cho KO. 2008. Prevalence and genotypes of pestivirus in Korean goats. *Korean J Vet Res* 48: 83-88.