The prevalence of causative agents of calf diarrhea in Korean native calves

Jeong-Byoung Chae¹, Hyeon-Cheol Kim², Jun-Gu Kang³, Kyoung-Seong Choi⁴, Joon-Seok Chae¹, Do-Hyeon Yu⁵, Bae-Keun Park⁶, Yeon-su Oh⁷, Hak-Jong Choi⁸* and Jinho Park⁸*

¹Laboratory of Veterinary Internal Medicine, BK21 PLUS Program for Creative Veterinary Science Research, Research Institute for Veterinary Science and College of Veterinary Medicine, Seoul National University, Seoul 08826, Korea
²College of Veterinary Medicine and Institute of Veterinary Science, Kangwon National University, Chuncheon 24341, Korea
³Korea Zoonosis Research Institute, Jeonbuk National University, Iksan 54531, Korea
⁴College of Ecology and Environmental Science, Kyungpook National University, Sangju 37224, Korea
⁵Institute of Animal Medicine, College of Veterinary Medicine, Gyeongsang National University, Jinju 52828, Korea
⁶College of Veterinary Medicine, Chungnam National University, Daejeon 34134, Korea
⁷Microbiology and Functionality Research Group, Research and Development Division, World Institute of Kimchi, Gwangju 61755, Korea
⁸Department of Veterinary Internal Medicine, College of Veterinary Medicine, Jeonbuk National University, Iksan 54596, Korea

Abstract
Infectious calf diarrhea is one of the most significant diseases of neonatal calves. This study is conducted to identify the prevalence of pathogens in calf diarrhea for 2 years. A total of 544 feces samples from Korean native beef calves were obtained to investigate selected seven pathogens causing calf diarrhea: bovine rotavirus, bovine coronavirus, Cryptosporidium parvum, bovine viral diarrhea virus, Eimeria species, Escherichia coli K99, and Salmonella species. The presence of diarrhea, the number and species of detected pathogens, and the calves’ ages were analyzed using various statistical methods depending on the case. Of the 7 target pathogens, 6 were detected in samples, but only Cryptosporidium parvum (p = 0.001) and bovine rotavirus (p < 0.001) were found at significantly higher rates in diarrheic calves than in non-diarrheic calves. Only Eimeria spp. showed a significant linear trend between the detection rate of the pathogen and the age groups (p < 0.05).

Keywords: Calf diarrhea, Korean native beef calves, Enteric pathogens, Prevalence

INTRODUCTION
Infectious calf diarrhea is one of the most significant diseases of neonatal calves. It has affected
the original work is properly cited.

**ORCID**

Jeong-Byung Chae  
https://orcid.org/0000-0003-3849-1063  
Hyeon-Cheol Kim  
https://orcid.org/0000-0002-8778-7277  
Jun-Gu Kang  
https://orcid.org/0000-0002-9083-8098  
Kyoung-Seong Choi  
https://orcid.org/0000-0002-2271-5360  
Joon-Seok Chae  
https://orcid.org/0000-0002-4813-3342  
Do-Hyeon Yu  
https://orcid.org/0000-0001-7645-6926  
Bae-Keun Park  
https://orcid.org/0000-0003-1241-6452  
Ye-on-su Oh  
https://orcid.org/0000-0001-5743-5396  
Hak-Jong Choi  
https://orcid.org/0000-0003-1185-0919  
Jinho Park  
https://orcid.org/0000-0001-5235-5717

**Competing interests**
The authors declare that they have no competing interests.

**Funding sources**
This research was supported by Technology Development Program (Project No. 116043-1) for Bio-industry, Ministry for Agriculture, Food and Rural Affairs, Korea and partially supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MSIT) (No. 2021R1A2C100517111) and by a research grant from the World Institute of Kimchi funded by the Ministry of Science and ICT, Korea (KE2101-1).

**Acknowledgements**
The authors would like to thank the owners of the cattle farms for cooperating with our research.

**Availability of data and material**
Upon reasonable request, the datasets of this study can be available from the corresponding author.

**Authors’ contributions**
Conceptualization: Choi KS, Chae JS, Yu DH, Park BK, Park J.  
Data curation: Chae JB, Kim HC.  
Formal analysis: Yu DH, Park BK, Oh Ys.  
Methodology: Kim HC, Choi KS, Park J.  
Software: Chae JB.  
Validation: Choi HJ, Park J.  
Investigation: Chae JB, Choi KS, Chae JS, Yu DH.  
Writing – original draft: Chae JB, Kang JG, Choi KS, Yu DH.  
Writing – review & editing: Oh Ys, Choi HJ, Park J.

**Ethics approval and consent to participate**
All procedures were performed according to ethical guidelines for the use of animal samples, as approved by Chonbuk National University (Institutional Animal Care and Use Committee [IACUC] Decision No. CBU 2016-0026).

In Korea, like other countries, calf diarrhea has had a serious impact on calf death. According to previous studies, 68.7% of calf deaths in Korean native beef calves and 53.4% in dairy calves were caused by digestive diseases [6,7]. Additionally, there have been several recent reports investigating pathogens that cause calf diarrhea [8–10]. However, most of them have been focused on specific pathogens from calf feces. As calf diarrhea can be caused by a variety of pathogens, it is necessary to simultaneously analyze different kinds of pathogens.

This study was performed to investigate the distribution of causative agents of calf diarrhea in Korean native beef calves aged less than 60 days in various regions of Korea and to discern their association with diarrhea.

**MATERIALS AND METHODS**

**Animals and sampling**
In this study, calves up to 60 days of age in 10 local Korean indigenous cattle farms in different areas of Korea (Yeongju, Samnye, Asan, Gimje, Mungyeong, Wanju, Heongseong, Sancheong, Iksan, Sangju) were selected for feces collection from 2016–2017. Feces were obtained by digital rectal palpation from the calves. All feces were scored as 0 to 3 using the scoring system included in the calf health scoring guide created by the University of Wisconsin–Madison School of Veterinary Medicine [11] and stored in 50 mL specimen bottles (SPL Life Sciences, Pocheon, Korea) at 4°C until they were transported to the laboratory. All feces scored at 2 and 3 were categorized as diarrhea.

**Pathogen detection**
All samples were examined for 7 pathogens (BCV, BRV, BVDV, *C. parvum*, *Eimeria* spp., *E. coli* K99, *Salmonella* spp.). Each feces sample was divided into two tubes and treated differently depending on the target agent, according to previously reported methods [8,12]. Briefly, to detect the 6 pathogens causing calf diarrhea (BCV, BRV, BVDV, *C. parvum*, *E. coli* K99, *Salmonella* spp.), fecal samples were suspended in 0.01 M phosphate-buffered saline to make 30% fecal homogenates and centrifuged for 1 min at 100×g. A supernatant was used to extract the total nucleic acid using MagMAX™ Total Nucleic Acid Isolation Kit (Thermo Fisher Scientific, Waltham, MA, USA). All extracts were stored at −70°C until real-time polymerase chain reaction (PCR) was performed. Real-time PCR was performed with the Path-ID™ Multiplex One-Step RT-PCR kit (Life Technologies, Carlsbad, CA, USA) according to the manufacturer’s recommended protocols in a 25 μL reaction volume using 8 μL of extracted template and 17 μL of the reaction mixture. Two types of real-time PCR were performed using specific primer sets for each pathogen in Table 1: one for the 3 viruses (BCV, BRV, BVDV) and the other for the bacteria and protozoa (*C. parvum*, *E. coli* K99, *Salmonella* spp.). Equal volumes of primers and probes were mixed for each target agent and the final concentration of each primer and probe was 0.2 μM. Real-time PCR was
Causative agents of calf diarrhea in Korea

performed using ABI 7500 Fast Real-Time PCR System (Applied Biosystems, Foster City, CA, USA). Cycling conditions of real-time PCR were as follows: (a) reverse transcription (RT) for 30 min at 45 °C; (b) activation of DNA polymerase for 10 min at 95 °C; (c) 40 cycles of denaturation at 94 °C for 15 sec and annealing/extension at 60 °C for 60 sec. RT step was performed only for viruses. After a 40 cycles reaction, samples with cycle threshold value less than 35 for targets were considered positive. To detect *Eimeria* spp., all fecal samples were suspended in a solution of 2.5% potassium dichromate and then transported to the laboratory. In the laboratory, fecal samples were analyzed to detect oocysts using the floatation methods with Sheather’s solution (saturated sugar solution; specific gravity 1.28) and examined microscopically (×400 magnification) based on the morphological features of the oocysts of the *Eimeria* spp.

**Table 1.** Nucleotide sequences of real-time polymerase chain reaction (PCR) primers and conditions for pathogens causing calf diarrhea

| Type                    | Microbial agents           | PCR primers, probes and conditions | Primer sequences (5' - 3') | Reference |
|-------------------------|----------------------------|------------------------------------|---------------------------|-----------|
| **Viruses** (PCR type 1)| Bovine viral diarrhea virus| BVD-F, BVD-R, BVD-Probe (CY5/BHQ2) | GGG NAG TCG TCA RTG GTT CG, GTC CCA TGT ACA GCA GAG WTT TT, CCA YGT GGA CGA GGG CAY GC | [23]      |
|                         | Bovine coronavirus         | BCV-F, BCV-R, BCV-Probe (FAM/MGB)  | CTA GTA ACC AGG CTA GTG TCA AAT CC, GGC GGA AAC CTA GTC GGA AAT, CCG CTG ACA TTC TCG ATC | [12]      |
|                         | Bovine rotavirus           | BRV-F, BRV-R, BRV-Probe (VIC/MGB)  | TCA ACA TGG ATG TCC TGT ATT CCT, TCC CCC AGT TTG GAA TTT ATT, TCA AAA ACT CTT AAA GAT GCA AG | [24]      |
| **Bacteria/parasites** (PCR type 2)| *Escherichia coli* K99 | K99-F, K99-R, K99-Probe (FAM/BHQ1) | GCT ATT AGT GGT CAT GGC ACT GTA G, TTT TTC TTC GCT AGG CAG TCA TTA, ATT TTAAC TAA AAC CAG CGCC CGG GCA | [25]      |
|                         | *Cryptosporidium parvum*  | *Cryptosporidium parvum*-F, *Cryptosporidium parvum*-R, *Cryptosporidium parvum*-Probe (JOE/BHQ1) | CAAATT GAT ACC GTT TGT CCT TCT GT, GGC ATG TCG ATT CTA ATT CAG CT, TGC CAT ACA TTG TTG TCC TGA CAA ATT GAA | [26]      |
|                         | *Salmonella species*       | *Salmonella*-F, *Salmonella*-R, *Salmonella*-Probe (CY5/BHQ2) | GGG NAG TCG TCA RTG GTT CG, GTG CCA TGT ACA GCA GAG WTT TT, CCA YGT GGA CGA GGG CAY GC | [27]      |

**Statistical analysis**

The PCR results for each pathogen were recorded as positive or negative and categorized based on diarrhea status and age group. Age group was divided into three age group 1 (1 d–10 d), age group 2 (11 d–30 d), and age group 3 (31 d–60 d). All statistical methods (The $\chi^2$, Fischer’s exact tests, and linear by linear association) were performed by SPSS v. 25.0 (IBM, Armonk, NY, USA). All graphical works were performed by GraphPad Prism 6 software (GraphPad, San Diego, CA, USA).
RESULTS

Relationship between fecal consistency and pathogen presence
Fecal samples collected from 544 Korean native beef calves on 10 local Korean indigenous cattle farms were described in Table 2. According to our results, diarrhea was not significantly associated with age group. The presence of pathogens in non-diarrheic calves was compared to that in diarrheic calves. Of 340 non-diarrheic calves, 213 calves (62.6%) were negative and 127 calves (37.4%) were positive for the pathogens examined. Alternatively, of 204 diarrheic calves, 101 calves (49.5%) were negative and 103 calves (50.5%) were positive for the pathogens. The presence of pathogens was significantly associated with diarrhea (odds ratio = 1.71, 95% confidence interval = 1.203–2.431, p < 0.01). And also there was a significant linear trend when comparing fecal scores and the number of detected agents (Fig. 1, p < 0.001).

The detection of 7 pathogens and relationship between diarrhea and each pathogen
The detection rate of the 7 pathogens in the normal feces and diarrheic feces is described in Table 3. *Eimeria* spp. (27.4%) was the most detected pathogen in overall samples, followed by BRV (8.8%), BCV (8.5%), *C. parvum* (4.4%), BVDV (0.7%), and *E. coli* K99 (0.2%). There was no *Salmonella* spp. in any of our samples. In the diarrheic samples, *Eimeria* spp. (31.4%) was detected most often, followed by BRV (15.2%), BCV (10.3%), *C. parvum* (8.3%), and *E. coli* K99 (0.5%). No BVDV or *Salmonella* spp. was detected. *C. parvum* (p = 0.001) and BRV (p < 0.001) had a significantly higher presence in diarrheic calves than in non-diarrheic calves.

Relationship between calves’ age and each pathogen
The detection rate of each pathogen according to age group was also compared (Fig. 2). *Eimeria* spp. was detected 33.3% (29/87), 29.5% (69/234), and 22.9% (51/223) in age group 1, 2, and 3, respectively. There was a significant linear trend between the detection rate of *Eimeria* spp. and the age group (p < 0.05). BRV was detected 6.9% (6/87), 10.7% (25/234), and 7.6% (17/223) in age group 1, 2, and 3, respectively. There was no significant linear trend between the detection rate of BRV and the age group. BCV was detected 6.9% (6/87), 8.5% (20/234), and 9.0% (20/223) in age group 1, 2, and 3, respectively. There was no significant linear trend between the detection rate of BCV and the age group. *C. parvum* was detected 6.9% (6/87), 3.8% (9/234), and 4.0% (9/223) in age group 1, 2, and 3, respectively. There was no significant linear trend between the detection rate of *C. parvum* and the age group. BVDV was detected 0% (0/87), 0.9% (2/234), and 0.9% (2/223) in age group 1, 2, and 3, respectively. There was no significant linear trend between the detection rate of BVDV and the age group. *E. coli* K99 was detected 0% (0/87), 0% (0/234), and 0.4% (1/223) in age group 1, 2, and 3, respectively. There was no significant linear trend between the detection rate of *E. coli* K99 and the age group.

DISCUSSION

In this study, the prevalence of the 7 pathogens in normal and diarrheic calves and the association between the pathogens causing calf diarrhea and the age and fecal status of 544 Korean native beef calves were demonstrated. As expected, diarrheic calves (50.5%) showed a significantly higher positive rate of pathogens than normal calves (37.4%), and the fecal consistency had a linear association with the number of detected pathogens, consistent with findings from other countries [13]. This suggested that pathogens were the one of the primary factors related to diarrhea in Korean native beef calves.
Three viruses (BRV, BCV, and BVDV) were detected in Korean native beef calves. BRV was detected 15.2% in Korean native beef calves and significantly related to diarrhea \((p < 0.001)\). In other reports in Korea, BRV was detected in 34.8% from diarrhea feces in Korean native calves [14], which might be come from the difference of regions, research periods, and methodology. However, these results including previous reports demonstrate that rotavirus is an important pathogen that can negatively affect the health of calves, consistent with that of earlier reports [13,15]. BCV was detected in non-diarrheic and diarrheic calves and there was no significant difference. Even though BCV is known as one of the main pathogens associated with calf diarrhea, this result that BCV were detected in normal feces was similar to that seen in earlier reports [3,16]. BVDV was detected in only 4 calves and all of them were in the non-diarrheic group. The detection rate of BVDV in this study was less than that in previous research [17]. This result might come from the type of

---

### Table 2. Description of calf feces collected

| Farm       | Age          | Normal | Fecal score | Diarrhea | Total |
|------------|--------------|--------|-------------|----------|-------|
|            |              | 0      | 1           | 2        | 3     | 4     |
|            |              | Subtotal | Subtotal | Subtotal |       |
| Total      | Age group 1  | 22     | 33          | 55       | 20    | 12    | 32    | 87    |
|            | Age group 2  | 59     | 82          | 141      | 57    | 36    | 93    | 234   |
|            | Age group 3  | 72     | 72          | 144      | 39    | 40    | 79    | 233   |
|            | Subtotal     | 153    | 187         | 340      | 116   | 88    | 204   | 544   |

---

### Table 3. Detection frequency of pathogens causing calf diarrhea from non-diarrheic and diarrheic feces of Korean native calves in Korea and association between a positive detection and calf diarrhea

| Pathogens          | Positive in overall samples | Positive in non-diarrheic calves | Positive in diarrheic calves | \(p\)-value | Odds ratio |
|--------------------|-----------------------------|----------------------------------|------------------------------|-------------|------------|
| Eimeria species    | 27.4% (149/544)             | 25.0% (85/340)                   | 31.4% (64/204)               | 0.113       | 1.37 (0.93–2.01) |
| Bovine rotavirus group A | 8.8% (48/544)               | 5.0% (17/340)                    | 15.2% (31/204)               | < 0.001     | 3.41 (1.83–6.33) |
| Bovine Coronavirus | 8.5% (46/544)               | 7.4% (25/340)                    | 10.3% (21/204)               | 0.286       | 1.45 (0.79–2.66) |
| Cryptosporidium parvum | 4.4% (24/544)               | 2.1% (7/340)                     | 8.3% (17/204)                | 0.001       | 4.33 (1.76–10.62) |
| BVDV               | 0.7% (4/544)                | 1.2% (4/340)                     | 0% (0/204)                   | 0.302       | 0.99 (0.98–1.00) |
| Escherichia coli K99 | 0.2% (1/544)                | 0% (0/340)                       | 0.5% (1/204)                 | 0.375       | 1.005 (1.00–1.02) |
| Salmonella species | 0% (0/544)                  | 0% (0/340)                       | 0% (0/204)                   | -           | -          |

1) Number in parentheses is the 95% confidence interval of the estimated odds ratio.
samples. Feces were used to detect BVDV in this research, however, ear notch, skin fold biopsies, and nasal swabs showed reliable results for the detection of BVDV than rectal swab [18].

Even though the detection rate of C. parvum was lower than that for Eimeria spp. and BCV, C. parvum was found at a significantly higher rate in diarrheic calves than in normal feces, similar to BRV. There have been many reports emphasizing the effects of C. parvum infection in calf diarrhea in other countries [3,13,19]. Because there is no worldwide commercial vaccine for C. parvum, maintaining good herd sanitation and keeping sick calves away from non-diarrheic calves are important in preventing C. parvum infections.

Two bacteria (E. coli K99 and Salmonella spp.) were selected in this research. There was only one calf positive for E. coli K99 in this research. This result was consistent with that of other reports in Korea that no E. coli strain expressing K99 was detected in isolated samples from cattle farms [20]. Salmonella spp. occurring calf diarrhea was not detected in this research. However, since Salmonella infection in other livestock and human have been reported in Korea [21,22], it is necessary to conduct ongoing monitoring of Salmonella infection in Korean beef calves.

In this study, Eimeria spp. was the most detected pathogen of the 7 examined pathogens and this detection rate was similar to that in other reports from Korea [8]. However, no significant difference was shown between non-diarrheic calves and diarrheic calves. Because Eimeria spp. was also detected frequently in the feces of non-diarrheic calves [23], this result was conceivable.

The amount of oocyte secretion was not investigated in this research, but the amount of oocytes excretion of Eimeria spp. is known to be strongly correlated with diarrhea, and thus, further research

### Table 4. The detection rate of Eimeria spp. from Korean native calves by age group and fecal status

| Fecal status | Age group | Eimeria spp. negative (%) | Eimeria spp. positive (%) | Total | p-value (linear for trend) |
|--------------|-----------|---------------------------|--------------------------|-------|---------------------------|
| Normal       | 1 (1–10)  | 36 (65.5)                 | 19 (34.5)                | 55    | 0.008                     |
|              | 2 (11–30) | 101 (71.6)                | 40 (28.4)                | 141   |                           |
|              | 3 (31–60) | 118 (81.9)                | 26 (18.1)                | 144   |                           |
|              | Total     | 255 (75.0)                | 85 (25.0)                | 340   |                           |
| Diarrhea     | 1 (1–10)  | 22 (68.8)                 | 10 (31.3)                | 32    | 0.956                     |
|              | 2 (11–30) | 64 (68.8)                 | 29 (31.2)                | 93    |                           |
|              | 3 (31–60) | 54 (68.4)                 | 25 (31.6)                | 79    |                           |
|              | Total     | 140 (68.6)                | 64 (31.4)                | 204   |                           |
should investigate the correlation between diarrhea in Korean native beef calves and the amount of *Eimeria* spp. excreted.

In comparing the age groups among calves to the pathogens detected, only *Eimeria* spp. showed a linear association to the age groups (Fig. 2). The prevalence of *Eimeria* infections in normal calves decreased as the age increased (*p < 0.01, linear trend), while in diarrheic calves, the prevalence was stable even as the age increased (Table 4). According to this result, ongoing investigations of the amount of *Eimeria* spp. infection are important in predicting the pattern of calf diarrhea by *Eimeria* species.

In conclusion, six of seven pathogens were detected in samples, but only *C. parvum* and bovine rotavirus were found at significantly higher rates in diarrheic feces than in non-diarrheic feces and *Eimeria* spp. showed a significant linear trend between the detection rate of the pathogen and the age groups.

**REFERENCES**

1. Cho YI, Yoon KJ. An overview of calf diarrhea - infectious etiology, diagnosis, and intervention. J Vet Sci. 2014;15:1-17. https://doi.org/10.4142/jvs.2014.15.1.1
2. Pereira RV, Adams-Progar AL, Moore DA. Dairy calf treatment for diarrhea: are the drugs we use effective? [Internet] Washington State University Extension. 2017 [cited 2021 Feb 12]. https://hdl.handle.net/2376/11916
3. Izzo MM, Kirkland PD, Mohler VL, Perkins NR, Gunn AA, House JK. Prevalence of major enteric pathogens in Australian dairy calves with diarrhoea. Aust Vet J. 2011;89:167-73. https://doi.org/10.1111/j.1751-0813.2011.00692.x
4. Peter SG, Gitau GK, Richards S, Vanleeuwen JA, Uehlinger F, Mulei CM, et al. Risk factors associated with Cryptosporidium, *Eimeria*, and diarrhea in smallholder dairy farms in Mukurwe-ini Sub-County, Nyeri County, Kenya. Vet World. 2016;9:811-9. https://doi.org/10.14202/vetworld.2016.811-819
5. Uhde FL, Kaufmann T, Sager H, Albini S, Zanoni R, Schelling E, et al. Prevalence of four enteropathogens in the faeces of young diarrhoeic dairy calves in Switzerland. Vet Rec. 2008;163:362-6. https://doi.org/10.1136/vr.163.12.362
6. Hur TY, Jung YH, Choe CY, Cho YI, Kang SJ, Lee HJ, et al. The dairy calf mortality: the causes of calf death during ten years at a large dairy farm in Korea. Korean J Vet Res. 2013;53:103-8. https://doi.org/10.14405/kjvr.2013.53.2.103
7. Kim UH, Jung YH, Choe C, Kang SJ, Chang SS, Cho SR, et al. Korean native calf mortality: the causes of calf death in a large breeding farm over a 10-year period. Korean J Vet Res. 2015;55:75-80. https://doi.org/10.14405/kjvr.2015.55.2.75
8. Kim HC, Choe C, Kim SH, Chae JS, Yu DH, Park J, et al. Epidemiological survey on *Eimeria* spp. Associated with diarrhea in Pre-weaned native Korean calves. Korean J Parasitol. 2018;56:619-23. https://doi.org/10.3347/kjp.2018.56.6.619
9. Lee SH, VanBik D, Kim HY, Lee YR, Kim JW, Chae M, et al. Multilocus typing of Cryptosporidium spp. in young calves with diarrhea in Korea. Vet Parasitol. 2016;229:81-9. https://doi.org/10.1016/j.vetpar.2016.09.019
10. Park J, Han DG, Kim SH, Chae JB, Chae JS, Yu DH, et al. Prevalence of coronavirus from diarrheic calves in the Republic of Korea. Asian Pac J Trop Biomed. 2018;8:1-6. https://doi.org/10.4103/2221-1691.221037
11. University of Wisconsin–Madison School of Veterinary Medicine. Calf health scoring chart [Internet]. 2015 [cited 2016 Feb 20]. https://fyi.extension.wisc.edu/heifermgmt/files/2015/02/calf_health_scoring_chart.pdf
Cho YI, Kim WI, Liu S, Kinyon JM, Yoon KJ. Development of a panel of multiplex real-time polymerase chain reaction assays for simultaneous detection of major agents causing calf diarrhea in feces. J Vet Diagn Invest. 2010;22:509-17. https://doi.org/10.1177/104063871002200403

Cho YI, Han JI, Wang C, Cooper V, Schwartz K, Engelken T, et al. Case-control study of microbiological etiology associated with calf diarrhea. Vet Microbiol. 2013;166:375-85. https://doi.org/10.1016/j.vetmic.2013.07.001

Lee SH, Kim HY, Choi EW, Kim D. Causative agents and epidemiology of diarrhea in Korean native calves. J Vet Sci. 2019;20:664. https://doi.org/10.4142/jvms.2019.20.664

Bartels CJM, Holzhauer M, Jorritsma R, Swart WAJM, Lam TJGM. Prevalence, prediction and risk factors of enteropathogens in normal and non-normal faeces of young Dutch dairy calves. Prev Vet Med. 2010;93:162-9. https://doi.org/10.1016/j.prevetmed.2009.09.020

Kirisawa R, Takeyama A, Koiwa M, Iwai H. Detection of bovine torovirus in fecal specimens of calves with diarrhea in Japan. J Vet Med Sci. 2007;69:471-6. https://doi.org/10.1292/jvms.69.471

Han DG, Ryu JH, Park J, Choi KS. Identification of a new bovine viral diarrhea virus subtype in the Republic of Korea. BMC Vet Res. 2018;14:233. https://doi.org/10.1186/s12917-018-1555-4

VanderLey B, Ridpath J, Sweiger S. Comparison of detection of bovine virus diarrhea virus antigen in various types of tissue and fluid samples collected from persistently infected cattle. J Vet Diagn Invest. 2011;23:84-6. https://doi.org/10.1177/104063871002300112

Trotz-Williams LA, Jarvie BD, Martin SW, Leslie KE, Peregrine AS. Prevalence of Cryptosporidium parvum infection in southwestern Ontario and its association with diarrhea in neonatal dairy calves. Can Vet J. 2005;46:349-51.

Shin SW, Byun JW, Jung M, Shin MK, Yoo HS. Antimicrobial resistance, virulence genes and PFGE-profiling of Escherichia coli isolates from South Korean cattle farms. J Microbiol. 2014;52:785-93. https://doi.org/10.1007/s12275-014-4166-1

Kang MS, Oh JY, Kwon YK, Lee DY, Jeong OM, Choi BK, et al. Public health significance of major genotypes of Salmonella enterica serovar Enteritidis present in both human and chicken isolates in Korea. Res Vet Sci. 2017;112:125-31. https://doi.org/10.1016/j.rvsc.2017.02.010

Oh SI, Kim JW, Chae M, Jung JA, So B, Kim B, et al. Characterization and antimicrobial resistance of Salmonella Typhimurium isolates from clinically diseased pigs in Korea. J Food Prot. 2016;79:1884-90. https://doi.org/10.4315/0362-028X.JFP-16-131

Gulliksen SM, Jør E, Løken T, Åkerstedt J, et al. Enteropathogens and risk factors for diarrhea in Norwegian dairy calves. J Dairy Sci. 2009;92:5057-66. https://doi.org/10.3168/jds.2009-2080

Mahlum CE, Haugerud S, Shivers JL, Rosso WD, Goyal SM, Collins JE, et al. Detection of bovine viral diarrhea virus by TaqMan® reverse transcription polymerase chain reaction. J Vet Diagn Invest. 2002;14:120-5. https://doi.org/10.1177/104063870201400205

West DM, Spriggins KA, Cassar C, Wakeley PR, Sawyer J, Davies RH. Rapid detection of Escherichia coli virulence factor genes using multiplex real-time TaqMan® PCR assays. Vet Microbiol. 2007;122:323-31. https://doi.org/10.1016/j.vetmic.2007.01.026

Guy RA, Payment P, Krull UJ, Horgen PA. Real-time PCR for quantification of Giardia and Cryptosporidium in environmental water samples and sewage. Appl Environ Microbiol. 2003;69:5178-85. https://doi.org/10.1128/AEM.69.9.5178-5185.2003

Moore MM, Feist MD. Real-time PCR method for Salmonella spp. targeting the stn gene. J Appl Microbiol. 2006;102:516-30. https://doi.org/10.1111/j.1365-2672.2006.03079.x